THE EFFECTS OF VARIOUS STABILIZERS ON THE MOUTHFEEL AND OTHER ATTRIBUTES OF DRINKABLE YOGURT

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The ingredients, processing conditions, and starter cultures used in producing a drinkable yogurt can affect the mouthfeel, viscosity, flavor, and other attributes. Chalkiness, a perceived defect of some dairy products, is an aspect of mouthfeel that is affected by the conditions associated with yogurt. The objective of this project was to determine how the mouthfeel and other attributes of drinkable yogurt would be altered by using gelatin, carboxymethylcellulose (CMC), and high methoxy pectin (HMP) as stabilizers. In addition, the related effects of heat treatment conditions and choice of starter cultures used in conjunction with these stabilizers were to be examined. Sensory analysis showed that the inclusion of the various stabilizers had effects on mouthfeel, viscosity, and flavor attributes. Yogurt samples produced with CMC, added prior to fermentation, were considered unacceptable and objectionable due to the inability to stabilize the yogurt matrix and negative effects on mouthfeel and taste. Gelatin and HMP produced the most viscous and chalkiest products, though the differences among the

products made with these stabilizers often depended on the heat treatment. However, none of the samples were considered objectionable with regard to chalkiness. The sensory data were correlated with pH, titratable acidity, soluble protein, and other measurements. The heating regimes (68.3 °C and 85 °C for 30 minutes) and starter cultures (ropy and nonropy) used were also found to influence the mouthfeel, texture, and flavor perceptions, although the effects differed, depending on the stabilizer. The starter cultures used affected the viscosity, mouth coating, sourness, pH, and titratable acidity, but the effect on overall acceptability was minimal. Overall, drinkable yogurts stabilized with gelatin or HMP were found to be the best overall for flavor and mouthfeel.

CHAPTER 1
INTRODUCTION

Yogurt is the result of the lactic acid fermentation of milk by two bacteria, L. *bulgaricus* and S. *thermophilus*. This dairy product is consumed worldwide and drinkable versions of yogurt have experienced steady growth in the United States during the past ten years. Higher customer demands and expectations create the necessity for maintaining the quality of yogurt.

Mouthfeel, flavor, and texture are important sensory aspects of yogurt quality. Research has shown that these attributes may be affected by the type of stabilizer used, heat treatment during processing, and the starter cultures selected for fermentation.

The application of heat to pasteurize milk causes unfolding of the serum (whey) protein peptide chains, resulting in denaturation of the proteins (Renner and Abd El-Salam, 1991; Strange *et al*., 1996; Walstra *et al*., 1999). Upon cooling, the denatured serum proteins associate and precipitate with casein, depending on pH (Fox and McSweeney, 1998). This is apparent in one of the main serum proteins, β-lactoglobulin, which associates with κ-casein on the casein micelle. The denatured serum proteins tend to aggregate, forming large, insoluble complexes that may increase the viscosity of the milk (Renner and Abd El-Salam, 1991; Walstra *et al.,* 1999).

The starter culture selected for the fermentation of milk to produce yogurt also affects the attributes of the yogurt. Strains of L. *bulgaricus* and S. *thermophilus* that produce exopolysaccharides are occasionally used in yogurt production. These "ropy" cultures secrete the polysaccharides as either a capsule or slime (Marth and Steele, 1998; Tamime and Robinson, 1999; Vedamuthu, 1991). The exopolysacharides, which consist primarily of glucose and galactose, but vary with the type of culture, crosslink with the bacterial cell surface and the protein of the yogurt matrix (Schellhaas and Morris, 1985; Tamime and Robinson, 1999). Although these bonds do not necessarily affect the firmness of the gel, the interactions contribute to an increase in viscosity and other rheological properties, which impact the overall mouthfeel of yogurt (Rawson and Marshall, 1998; Schellhaas and Morris, 1985).

Stabilizers may also be added to alter or improve the characteristics of a yogurt system. Gelatin, a protein-derived hydrocolloid, forms junction zones as amino acids in the gelatin structure change conformation upon heating and cooling (Imeson, 1997). Hydrogen bonds stabilize the structure, resulting in a gel (Harris, 1990; Phillips and Williams, 2000). The strength of the gel and other properties like viscosity and melting point (which imparts mouthfeel) may depend on the conditions of the system. Concentration, temperature, and pH are particularly important. These conditions also affect the use of other hydrocolloids, such as carboxymethylcellulose (CMC). In dairy products, this polysaccharide stabilizer produces heat-stable, soluble complexes through reactions with casein at the isoelectric point (Imeson, 1997; Phillips and Williams, 2000). Hence, CMC may find practical use for yogurt drinks (Tamime and Robinson, 1999). Pectins, such as high- and low- methoxy pectins (HMP and LMP, respectively), may also

be used for dairy products. While LMP gels in the presence of calcium ions, HMP gels require sugar and acid (Harris, 1990; Imeson, 1997). The various conditions in the food system and the effects of the hydrocolloids on properties like mouthfeel and viscosity are considerations in stabilizer selection for the production of drinkable yogurt.

Mouthfeel and texture are important attributes for yogurt acceptance by consumers (Barnes *et al*., 1991; Brennan *et al*., 2002). Chalkiness, considered a defect in mouthfeel of some dairy systems, may be influenced by factors such as processing conditions, stabilizers, and starter culture selection (Maiolino, 2002; Wszolek *et al*., 2001). The objective of this research is to investigate the effects of gelatin, CMC, and HMP on the mouthfeel, particularly related to chalkiness, of a drinkable yogurt system. As processing conditions often vary, the drinkable yogurt produced for these experiments will be heated to two different temperatures and both ropy and nonropy starter cultures will be used.

CHAPTER 2
LITERATURE REVIEW

Although drinkable forms of yogurt and related cultured dairy products have been popular in Europe and other international markets for a considerable time, the popularity of this product has only gained acceptance with consumers in the United States in the past decade. As an industry estimated to have approximately $12.6 million in sales in 1997 (Author Unknown, 2002), the cultured dairy beverages category increased more than 250% over the following four years (Author Unknown, 2002; Sloan 2002). In 2001, sales of these products, which include drinkable yogurt, yogurt juice drinks, smoothies, and kefir, reached $86.2 million and marketing research expects this area to grow 20.7% from 2001 to 2006 (Author Unknown, 2002).

The dramatic increases in sales and popularity of drinkable yogurt and related products may be attributed to the overall image of a yogurt beverage as a healthy, convenient, and satisfying food choice. Thus, the importance of the flavor, texture, and mouthfeel cannot be overlooked. These factors are largely dependent upon various aspects of processing, including, but not limited to, temperature effects, ingredient choices, and starter culture selection.

Yogurt Processing

Although there are no standardized procedures for making a drinkable yogurt product, most processors agree on a general process. This includes pre-treatment of the milk, heat treatment, homogenization, cooling, started culture addition and subsequent fermentation, and packaging (Tamime and Robinson, 1999).

Pre-treatment

The pre-treatment step involves adjusting the milk before processing. In set or stirred yogurt production, this may include the addition of milk solids to achieve desired viscosity. However, for a drinkable yogurt, milk without fortification is normally used (Tamime and Robinson, 1999). Pre-treatment also includes the standardization of the fat content in the milk. Standardization of the fat is an important step, not only for adherence to legal standards, but also for the overall effect on the flavor and texture attributes of the finished product, particularly relating to the viscosity and mouthfeel. According to Tamime and Robinson (1999), milk can be standardized in a variety of ways. Part of the fat content may be removed from the milk, full cream milk may be mixed with skimmed milk, cream can be added to full fat or skimmed milk, or any combination of these using standardizing centrifuges.

The pre-treatment of the milk also includes the addition of any stabilizers. Whey separation, a typical quality defect in drinkable forms of yogurt, may be prevented by the inclusion of various stabilizers such as gelatin (Tamime and Robinson, 1999).

Heat Treatment

The application of heat is a crucial step in yogurt production and affects both the safety and quality of the product with both temperature and time being critical factors. The primary reason for this heat treatment is the destruction of most of the pathogenic or potentially pathogenic microorganisms associated with milk (Early, 1998; Tamime and Robinson, 1999; Walstra *et al.*, 1999). However, typical pasteurization does not necessarily include the destruction of bacterial spores or certain enzymes.

Originally a batch process, the pasteurization of milk encompasses a wide range of acceptable temperatures and times. For instance, a low temperature, long time process

would involve processing the milk at 145 °F (62.8 °C) for 30 minutes. A high temperature, short time production, such as commonly used in continuous processing, would involve the application of temperatures at 161 °F (71.7 °C) for 15 seconds In addition to the pasteurization of the milk, these conditions also affect the properties of the milk for the yogurt, as well as for the starter cultures (Labropoulous *et al.*, 1980; Tamime and Robinson, 1999; Walstra *et al.*, 1999).

Much research has been conducted on the physical and chemical changes in milk associated with heating regimes. Flavor and viscosity changes are only a few of the many alterations. A slight increase in viscosity resulting from heat treatment may be attributed to the effects of the heat on the serum proteins in the milk. Often referred to as whey proteins, serum proteins consist mainly of the globular proteins β-lactoglobulin, ∝-lactalbumin, serum albumin, and various immunoglobulins (Walstra *et al.*, 1999). High temperatures (i.e. 70 – 90 °C) cause the peptide chains of these proteins to unfold, resulting in denaturation of the protein and increased viscosity (Renner and Abd El-Salam, 1991; Strange *et al.*, 1996; Walstra *et al.*, 1999). Depending on the heating temperature, such as those used in high-temperature pasteurization, and the pH, various reactions on the side chains of these proteins may take place, preventing the peptide chain from reverting back to the original conformation. These denatured proteins are insoluble (Walstra *et al.*, 1999).

A variety of reactions involving the serum proteins may occur, but these are largely dependant upon the pH. For instance, application of heat at high temperatures (greater than 80 °C) causes β-lactoglobulin to associate with κ-casein on the outer layer of the casein micelle (Fox and McSweeney, 1998). As a result of these types of reactions, the

volume of the proteins increase, resulting in a slight increase in milk viscosity (Renner and Abd El-Salam, 1991).

Heating and cooling the milk, followed by acidification to the isoelectric pH of casein, as is done in the production of yogurt, causes the denatured serum proteins to associate and precipitate with the casein (Fox and McSweeney, 1998). Walstra *et al.* (1999) indicates that other reactions may occur that cause dimers and other small, soluble aggregates of protein to be formed. Depending upon pH and other conditions, these may continue to aggregate. These larger complexes of denatured serum proteins are insoluble. Proteose-peptone is the only serum protein to which denaturation does not apply since this protein is not globular (Renner and Abd El-Salam, 1991; Walstra *et al.*, 1999).

Similarly, casein does not denature. However, very high heat treatments, such as those used for sterilization of concentrated milk, can cause the casein to irreversibly aggregate. Again, depending primarily on pH (generally below pH 6.2), the large aggregates, bonded by covalent crosslinking, coagulate and a gel may be formed (Fox and McSweeney, 1998; Walstra *et al.*, 1999). Other reactions and conditions, such as those involving calcium bridging or the depletion of κ-casein from the micelles, may play a role in the heat coagulation. Although this aggregation of casein and possible subsequent coagulation may affect the viscosity of the milk more than the denaturation of the serum proteins, very high temperatures and other specific conditions must be met. Hence, these reactions do not typically occur in the regular processing of milk for yogurt (Fox and McSweeney, 1998; Walstra *et al.*, 1999).

In addition to the effects on viscosity, the application of heat has many other effects on milk. Flavor changes due to Maillard reactions, formation of free sulfhydral groups,

protein interactions, and lactone and methyl ketone formation are apparent (Fox and McSweeney, 1998; Walstra *et al.*, 1999). Furthermore, color, nutrients, and many other attributes of milk are affected (Robinson, 1994).

The gel matrix of yogurt has been examined by scanning electron microscopy (SEM). In heated milk, chains and clusters of casein micelles are formed, which serves to immobilize the liquid phase (Kalab, 1981). This also results in protein being evenly distributed, producing a firmer and more stable yogurt (Tamime and Robinson, 1999). However, in yogurt made from unheated milk, this does not occur as the casein micelles fuse into larger aggregates that are not distributed evenly in the matrix. As a result, a weaker yogurt is produced and is more susceptible to syneresis (Kalab, 1981; Tamime and Robinson, 1999). According to Tamime and Robinson (1999), the porosity of the protein matrix is impacted by the amount of casein and non-casein protein in the milk. SEM revealed that the yogurt is more susceptible to whey separation with larger pores in the yogurt matrix (Kalab, 1981; Tamime and Robinson, 1999). Electron microscopy was also used to examine the effects of some stabilizers on the yogurt gel matrix (Kalab, 1981). Ingredients such as carageenan caused aggregation of casein or the formation of fibers from the stabilizer to the casein micelles (Kalab, 1981). Gelatin, however, could not be examined by the electron microscopy techniques employed (Kalab, 1981).

SEM was also used to analyze the effects of the starter cultures on the gel matrix of the yogurt. This revealed that the lactic acid bacteria used in yogurt production formed "pockets" in the gel structure as a result of bacterial action (Kalab, 1981, Tamime and Robinson, 1999). Further study of exopolysaccharide-producing starter cultures showed that filaments of the polysaccharides produced are formed. These filaments attach to the

protein of the yogurt coagulum, which possibly improves the viscosity and stability of the gel (Kalab, 1981; Modler and Kalab, 1993; Tamime and Robinson, 1999).

Homogenization

The next step in yogurt processing also impacts the physical and chemical aspects of the milk. Homogenization breaks the large fat globules into smaller ones, thereby creating a stable emulsion out of an oil-in-water mixture (Early, 1998; Spreer, 1998). This is typically accomplished by directing the standardized milk through small valves at high pressure (Tamime and Robinson, 1999). This action reduces the size of the fat globules, which may be from 1 to 18 μm in diameter (average size is 3-4 μm) to less than 2 μm (Early, 1998; Tamime and Robinson, 1999). The new fat globules are stabilized by binding to some of the casein that was broken during this processing step (Fox and McSweeney, 1998; Tamime and Robinson, 1999). As a result, homogenization serves several purposes, including preventing the separation of the cream layer, improving stability, altering the physical attributes, etc. (Tamime and Robinson, 1999; Walstra *et al.*, 1999). These effects are dependant upon the amount of fat in the milk, as well as the pressure and temperature used for homogenization and may be minimal in skim milk products (Fox and McSweeney, 1998; Tamime and Robinson, 1999). In addition, some references indicate that this processing step should occur prior to the heat treatment (Tamime and Robinson, 1999). Regardless, both the serum proteins and the fat in the milk may be affected by homogenization. Some denaturation of these proteins, primarily by β-lactoglobulin and ∝-lactalbumin, may occur along with the resultant effects as previously discussed with the heat treatment (Tamime and Robinson, 1999). Heating the homogenized milk at a temperature above 70 °C may induce the denatured serum

proteins to interact with other denatured β-lactoglobulin to form a gel. Interactions with κ-casein both on the surface of suspended micelles and adsorbed on the fat globules are possible, as are interactions with the residual fat globule membrane. Adsorbed casein can also be displaced on the fat globule by adsorbing onto the globule surface (Tamime & Robinson, 1999). The reduction in size of the fat globules and increase in the adsorption on the casein micelle leads to an increase in total volume, resulting in an increased viscosity and the formation of a softer gel (Spreer, 1998; Tamime and Robinson, 1999). Color is also affected, with homogenized milk appearing whiter due to the scattering and reflectance of light by a larger number of fat globules (Fox and McSweeney, 1998; Tamime and Robinson, 1999). The interaction between the casein and the fat globules, as well as other protein effects, result in an increase in hydrophobicity and water binding which limits syneresis (Tamime and Robinson, 1999). Protein stability may also be decreased from the various protein interactions.

Starter Cultures

The starter cultures, *L. bulgaricus* and *S. thermophilus,* are required for the fermentation in yogurt production (Vedamuthu, 1991). For this reason, both the interaction of the cultures and their individual characteristics are important for flavor and texture development.

L. bulgaricus is a lactic acid bacteria that has been found to be relatively sensitive to low temperatures. According to Vedamuthu (1991), this microorganism is usually seen as rod shaped and, although typically slender, may be curved or in pairs or chains. As a homofermentative thermophile, *L. bulgaricus* is known to be relatively tolerant to heat, with an optimum growth temperature of approximately 45-50 °C (Rasmussen, 1981;

Rybka and Kailasapathy, 1995; Vedamuthu, 1991). As a result of this high temperature resistancy, this yogurt starter microorganism is commonly used in high temperature fermentations (Rasmussen, 1981). The association of *L. bulgaricus* with other lactobacilli is under debate (Tamime and Deeth, 1980). Certain characteristics, however, serve to distinguish these bacteria from other members in the family Lactobacillacae, such as *L. helveticus*, which is also found in some yogurt and cheeses (Rasmussen, 1981). As described by Rasmussen (1981), these differences include the production of granules by *L. bulgaricus*, as seen under a microscope, an attribute which *L. helveticus* lacks. A second distinguishing feature is the maximum titratable acidity production of approximately 1.7% for *L. bulgaricus*. This is considerably lower than *L. helveticus*, which maintains a maximum titratable acidity for acid production in milk of about 2.7%. A third characteristic of *L. bulgaricus* as compared to *L. helveticus* is the inability of *L. bulgaricus* to ferment maltose (Rasmussen, 1981). *L. bulgaricus* is considered to be a relatively slow-growing organism, due to a lack of strong tolerance for oxygen. As a result of this, until the oxygen levels are reduced during fermentation, this microorganism will not grow rapidly (Marth and Steele, 1998).

The other starter microorganism involved in yogurt production, *S. thermophilus*, is spherical-shaped, typically found in pairs or long chains, and is noted for the ability to withstand higher temperatures, contributing to the classification of homofermentative thermophile (Rasmussen, 1981; Tamime and Deeth, 1980; Vedamuthu, 1991). The thermal resistance is demonstrated in the evidence of *S. thermophilus* survival during heating at 60 °C for 30 minutes and characterized by the use of these bacteria in various high temperature fermentations (Rasmussen, 1981; Tinson *et al.*, 1982; Wilkins, *et al.*

1986). Despite being able to survive at higher temperatures, *S. thermophilus* fails to grow at 10 °C, separating this microorganism from other streptococci that grow at lower temperatures (Marranzini *et al.*, 1987; Tamime and Deeth, 1980). Other apparent attributes aside from optimum, minimum, and maximum growth temperatures, include the inability of *S. thermophilus* to grow in high salt concentrations and the inability to produce ammonia from arginine (Marth and Steele, 1998). These attributes are important for identification, since *S. thermophilus* does not possess a necessary antigen for serological identification and, therefore, physiological techniques must be utilized (Marranzini, *et al.*, 1989; Tamime and Deeth, 1980). Other characteristics of this member of the Streptococceace family include a sensitivity to phosphates and, as is the case with *L. bulgaricus*, production of capsules and exopolysaccharides by some strains. Much research has been conducted on the capsule material in terms of the effects on viscosity and texture.

S. thermophilus and *L. bulgaricus* exist in a complex cooperative relationship in yogurt, in which one bacterium produces stimulatory agents for the other (Schmidt, *et al.*, 1989; Tamime and Deeth, 1980; Wilkins *et al.*, 1986; Vedamuthu, 1991). *L. bulgaricus* has been shown to produce certain amino acids, such as valine, leucine, and histidine, which are essential for *S. thermophilus* to grow. These amino acids are the result of proteolysis of casein by *L. bulgaricus* (Marranzini *et al.*, 1987; Abu-Tarboush, 1996). Numerous additional studies have been performed to determine which amino acids stimulate *S. thermophilus* under varying conditions. However, according to Abu-Tarboush (1996), *S. thermophilus* is stimulated by valine, histidine, methionine, glutamic acid, and leucine released by *L. bulgaricus*. In reciprocation, *S. thermophilus* stimulates

L. bulgaricus by the production of various compounds under anaerobic conditions (Abu-Tarboush, 1996; Tamime and Deeth, 1980). As listed in a study by Abu-Tarboush (1996), several of these substances include peptides, purines, pyrimidines, oxaloacetic acid, and fumaric acids, though Tamime and Deeth (1980) cite previous research that contends that peptides, purines, and pyrimidines may not necessarily stimulate growth of *L. bulgaricus*. In addition to these compounds, *S. thermophilus* produces formic acids and carbon dioxide, and reduces the oxygen-reduction potential, which serves to increase the acid produced by *L. bulgaricus* (Marranzini, *et al.* 1989). As both *L. bulgaricus* and *S. thermophilus* partially convert the lactose in the milk to lactic acid, this is an important aspect of the carbohydrate fermentation during yogurt production, as is the production of acetaldehyde by these microorganisms (Labell, 1989). As a result of both lactic acid and acetaldehyde generation from this cooperative growth, the characteristic flavor of yogurt is produced (Marranzini *et al.*, 1989; Rasmussen, 1981; Richter and Mull, 1975; Schmidt *et al.,* 1989; Wilkins *et al.,* 1986a; Wilkins *et al.*, 1986b).

Both *L. bulgaricus* and *S. thermophilus*, as well as other lactic acid bacteria, have "ropy" strains that produce exopolysaccharides, either as a capsule or secreted as slime (Marth and Steele, 1998, Tamime and Robinson, 1999, Vedamuthu, 1991, Wood and Holzapfel, 1995). However, Hassan *et al* (1996a, 1996b) determined that several strains of encapsulated lactic acid bacteria could not be characterized as ropy and, therefore, differentiate between encapsulated ropy, encapsulated nonropy, and unencapsulated nonropy strains. In other research, no distinction is necessarily made between the versions of polysaccharide secretions. Regardless, these strains have been studied for their possible benefit of the polysaccharide production to the viscosity, mouthfeel, and

other rheological properties. Water-binding capacity of yogurts made with these bacteria related to the syneresis or "wheying off" defect in set and stirred yogurt have also been examined (Tamime and Robinson, 1999).

A study conducted by Schellhaas (1983) indicated that the apparent viscosity of skim milk gels made by ropy cultures was increased compared to that made by non-ropy cultures (Tamime and Robinson, 1999). Further experimentation by Schellhaas and Morris (1985) maintained that, at higher shear rates, gels produced by ropy cultures exhibit less of a decrease in viscosity. Electron microscopy revealed that this might be due to the crosslinking of the exopolysaccharide with both the cell surface and the protein of the gel (Schellhaas and Morris, 1985). However, as a result of this bonding, gel firmness is not necessarily improved by the presence of these exopolysaccharides.

Rawson and Marshall (1997) compared the effects of ropy and nonropy yogurt cultures on the viscosity and gel texture of stirred yogurt. Results indicated that apparent viscosity was increased by using both ropy strains of *L. bulgaricus* and *S. thermophilus* as compared to yogurt made with nonropy cultures. However, after destructive shear rate-thinning and subsequent recovery time, viscosity was not recovered as well for yogurt made with the two ropy cultures. Rheological data from this study also suggests that although ropiness may improve cohesiveness, firmness (defined as "the force necessary to attain a given deformation") of the yogurt matrix may not be affected. These results correlate to previous research (Schellhaas and Morris, 1985), where the increase in viscosity was attributed to a shorter relaxation time after shearing. This suggests that the polysaccharide-protein association might have more of an impact on the viscosity than the protein-protein interactions of nonropy yogurt (Rawson and Marshall, 1997; Tamime

and Robinson, 1999). Other researchers determined that these bonds are stretched under low shear rates but are broken with higher shearing (Rawson and Marshall, 1997). According to Rawson and Marshall (1997), the reduction in firmness may result from the excess slime produced prohibiting the polysaccharide-protein bonds from being formed, resulting in a weaker structure of the gel (Rawson and Marshall, 1997). The researchers concluded that the relaxation time of the bonds do not affect the gel firmness. The polysaccharide-protein interactions from ropy bacteria do, however, contribute to the overall viscosity, cohesiveness, and adhesiveness, which may have an influence on the overall mouthfeel of the yogurt (Rawson and Marshall, 1997).

Hassan *et al.* (1996a; 1996b) examined the rheological and textural properties of yogurt made with strains differentiated as encapsulated ropy (in which an exopolysaccharide capsule is formed), encapsulated nonropy (secretes extracellular slime), and unencapsulated nonropy. Yogurts made with ropy cultures exhibited increased viscosity and shear stress values; however, differences attributed to the type of polysaccharide secretion (capsule or slime) were apparent. Results from these experiments showed that the presence of bacterial capsules may enhance some rheological properties, such as viscosity, but may weaken the gel structure. This caused lower shear stress values compared to slime-producers, which produce a more stretchable gel structure (Hassan *et al.*, 1996a). The size of the capsule also affected the gel structure. Larger capsules created structures with large pores, which further served to enhance viscosity (Hassan *et al.*, 1996a). Overall, encapsulated nonropy cultures produced yogurt that were more stable for shearing, were more viscous, and had a lower yield stress (Hassan *et al.*, 1996a; Marth and Steele, 1998).

The type of extracellular polysaccharide produced by the yogurt bacteria was shown to have effects on the texture and syneresis, as well. Yogurt made with encapsulated nonropy cultures had the lowest firmness and curd tension, but exhibited less syneresis than unencapsulated cultures (Hassan *et al.*, 1996b). The lower firmness in the yogurt made by slime producing cultures may have been due to the polysaccharide interfering with the casein structure (Hassan *et al.*, 1996b). Hassan *et al.* (1996b) also demonstrated that, in addition to the type of polysaccharide produced, yogurt texture was affected by pH, whether a single strain or a mixed culture was used, and size of the capsular material produced.

Production of exopolysaccharides by starter cultures influences the flavor intensity of yogurt. High polysaccharide-producing cultures may impart a higher intensity due to the carbohydrate masking the flavor (Tamime and Robinson, 1999). Mouthfeel and other attributes may be affected as well.

Stabilizers

Stabilizers have been used in food products for a variety of purposes, including thickening, aiding stability, and improving mouthfeel (Harris, 1990; Imeson, 1997; Phillips and Williams, 2000). According to Glicksman (1982), stabilizers can be categorized according to the manufacturing process (Tamime and Robinson, 1999). Hydrocolloids can either be of natural, modified, or synthetic origin. Selection of the stabilizer or stabilizer combination to be used in a food system greatly depends on several variables. Functional properties of the stabilizer, intended use and outcome, interactions with other ingredients, and legal aspects are only a few considerations. In yogurt, stabilizers are added for two main reasons – as thickening or gelling agents and to stabilize the yogurt matrix (Early, 1998; Phillips and Williams, 2000; Tamime and

Robinson, 1999). In this capacity, the hydrocolloids, which are generally added to the milk prior to fermentation, can improve the viscosity, maintain the yogurt structure, inhibit syneresis, alter the mouthfeel, and, in the case of yogurt with added fruit, help keep the fruit in suspension (Early, 1998). Gelatin, carboxymethylcellulose, and high methoxy pectin all may be used to achieve these results.

Gelatin is a natural stabilizer derived from collagen (Harris, 1990; Imeson, 1997), usually from pigskin, cattle hides, or cattle bones (called ossein). The conversion from this animal protein into gelatin is accomplished by either an acid or alkali hydrolysis pre-treatment. The acid process involves soaking the pigskins or bones in a 5% maximum acid bath, usually of hydrochloric, sulfuric, or phosphoric acid, for 10-48 hours (Harris, 1990; Imeson, 1997). Unlike the previous treatment, the basic process is much longer, ranging from 6 to 20 weeks. This requires treating the ossein or hides with an alkali, typically lime (Harris, 1990; Imeson, 1997). The goal of both the acid and alkali treatments is to break the chemical cross-linkages in the fibers of the collagen, thereby creating a product that is soluble in water (Harris, 1990; Imeson, 1997). Gelatin made from the acid pre-treatment is categorized as Type A, while Type B gelatin results from the alkali processing. These two types differ primarily in the isoelectric points, ranging from 6.5 to 9.4 (6.5 – 7.5 for ossein gelatin, 7.5-9.0 for pigskin) for Types A and B, respectively (Harris, 1990; Imeson, 1997; Phillips and Williams, 2000). These differences have been attributed to side reactions during pre-treatment, including the hydrolysis of asparagines and glutamine to release ammonia in the alkali treatment (Harris, 1990; Phillips and Williams, 2000). Side-group reactions are less severe in the acid treated gelatin, resulting in the isoelectric range being closer to that of collagen (pH

9.4) (Harris, 1990; Imeson, 1997). The differences in the isoelectric points affect the usage of the gelatin. For instance, Type B gelatin is recommended for blending with negatively charged polysaccharides like carageenan (Harris, 1990; Imeson, 1997).

Gelatin is comprised mostly of amino acids held together with peptide bonds in a left-hand helix with no covalent crosslinking. Proline, hydroxyproline, and glycine are predominant, though nearly all amino acids are found. The exception is tryptophan, though cysteine is only present in minute quantities (Imeson, 1997). The gelatin is melted upon the application of heat due to the relatively low melting point (27 °C to 34 °C). According to Phillips and Williams (2000), the portions of the gelatin chain that contain pyrolidine are important for forming junction zones. Upon cooling, those areas, particularly those that contain glycine-proline-proline sequences contain a left-hand turn of the helix structure. Aggregation of three of these helices creates junction points, similar to those found in collagen and hydrogen bonds stabilize the structure (Fennema, 1994; Harris, 1990; Imeson, 1997; Phillips and Williams, 2000). The result is the formation of a three-dimensional gel structure (Imeson, 1997).

The properties of this gel are very important in terms of the application for food use. One attribute to be considered is the gel strength, also referred to as the bloom or the bloom strength. This is defined as the weight in grams needed to produce a four-millimeter deep depression by a piston (12.7 mm in diameter) in the surface of a gel (6.67% concentration that has been set for 16 to 18 hours at 10 °C) (Imeson, 1997; Phillips and Williams, 2000). The strength of these gels can be affected by several factors, including pH, temperature, setting time, and interactions with other ingredients.

Similarly, these factors may also affect other characteristics of a gelatin gel. The melting point, for example, may be altered by the concentration, setting conditions, and other ingredients (Harris, 1990; Imeson, 1997; Phillips and Williams, 2000). Likewise, viscosity may be affected, particularly by temperature, pH, and concentration. These effects on the melting point and viscosity may influence the mouthfeel and other sensory characteristics.

Carboxymethylcellulose (CMC) is a polysaccharide hydrocolloid compound of β 1-4 glucose polymers that have been derivitized with carboxy methyl groups. Derived from cellulose, the properties of CMC can vary, depending on the molecular weight, degree of polymerization, degree of substitution, ingredient interactions, and other conditions (Imeson, 1997). Molecular weight and degree of polymerization, for instance, can produce CMC that ranges from low to high viscosities given a certain concentration in solution. In addition, the viscosity of CMC solutions is inversely related to temperature, though Imeson (1997) cautions that prolonged exposure to high temperatures can irreversibly degrade CMC, which affects the viscosity upon cooling. Viscosity may also be affected by pH, depending on the range. Acidic conditions, such as those below pH 3.0, result in an increased viscosity and precipitation of the free acid form of CMC, so use of this hydrocolloid is not recommended for these products (Imeson, 1997).

Rheological aspects, however, appear to be affected by the degree and uniformity of anhydroglucose substitution along the modified cellulose chain (Phillips and Williams, 2000). Random substitutions allow the anhydroglucose units to form a three-dimensional, thixotropic matrix after shearing (Imeson, 1997). Uniform substitution

aligns these units after shearing, shifting the flow from a thixotropic system to a smoother flow (Imeson, 1997).

CMC tends to be used as a stabilizer in dairy products, as this hydrocolloid can react with casein near the isoelectric point, producing a soluble complex (Imeson, 1997). According to Phillips and Williams (2000), CMC/casein complexes are stable under heating, with slight decreases in viscosity. However, this system is affected by shearing and agitating decreases the viscosity. As a result, this polymer finds possible use in yogurt-based drinks at a recommended usage level of 0.5% at pH 4.3-4.4 (Imeson, 1997, Tamime and Robinson, 1999). These conditions require the stabilizer to be added after fermentation. If added prior to fermentation, protein agglomeration, whey separation, and other defects are more likely (Imeson, 1997).

Pectins, although typically associated with fruit jellies and preserves, also can be utilized for yogurt production. These gelling agents are derived from citrus peels and apple pomace (Harris, 1990; Imeson, 1997). Although the chemistry of pectin has not been fully elucidated, pectin is generally thought to be comprised of 1,4-linked ∝-D-galacturonic acid (Harris, 1990; Imeson, 1997; Williams and Phillips, 1998). Up to 80% of these groups may be methanol-esterified (Harris, 1990; Phillips and Williams, 2000). By definition, high-methoxy pectin (also referred to as high-methoxyl pectin or high-ester pectin), denoted HMP, is greater than 50% esterified. Conversely, low-methoxy (low-methoxyl or low-ester; LMP) pectins have less than 50% methyl-esterification of the galacturonic subunits (Harris, 1990; Imeson, 1997; Phillips and Williams, 2000). These variations in degree of esterification influence the properties of commercial pectins. In the presence of calcium ions, low methoxy pectins gel by forming structures referred to

as “egg boxes” (Harris, 1990; Phillips and Williams, 2000; Williams and Phillips, 1998). Although these were originally described for alginates, these structures result from the calcium ions creating junction zones between the carboxylic acids on different molecules. The large number of methoxy groups associated with HMP mostly inhibits gelling in the presence of calcium (Harris, 1990; Imeson, 1997). Instead, these highly esterified pectins can gel with sugar and acid, a characteristic not shared with LMP. The acid environment causes the side chains to become protonated while the sugar molecules dehydrate the structure, creating junction zones, which form the gel. The many non-esterified carboxylic acids in LMP prevent the structure from being dehydrated enough to gel. Hence, HMP are used in gelling applications with higher sugar contents (Harris, 1990; Imeson, 1997; Phillips and Williams, 2000). According to Phillips and Williams (2000), HMP is often used as a stabilizer for acid products, such as drinkable yogurts. This thermally irreversible hydrocolloid can be added to a milk product at higher pH, then immediately acidified. The result is a product with a gel strength that is maintained to low pH values (Phillips and Williams, 2000). In this capacity, HMP is useful by providing a drinkable yogurt that exhibits good mouthfeel characteristics, is not chalky, and does not sediment (Phillips and Williams, 2000).

These two types of pectin also differ in setting. Low methoxy pectin will set almost as soon as appropriate conditions are met. High methoxy pectins, however, are categorized as slow-set, medium-set, and rapid-set (Harris, 1990; Imeson, 1997; Phillips and Williams, 2000). In general, the higher the degree of esterification, the faster the gel is set.

Gelatin, CMC, and HMP, as well as other types of stabilizers, have been studied for their effects on yogurt and yogurt products. Research by Towler (1984) examined the effects of propylene glycol alginate (PGA), CMC, and pectin on the viscosity and sedimentation of a cultured milk beverage. Viscosity increased with the addition of up to 2% (w/v) stabilizer, then decreased until 0.4% (w/v) stabilizer was reached. Larger amounts of stabilizers added resulted in a rapid increase in viscosity. Sedimentation of the milk protein increased with lower levels of stabilizers, but decreased once the level of stabilizer "increased beyond the level of minimum viscosity" (Towler, 1984). Irregardless of quantity, products made with CMC sedimented greatly, so PGA and pectin were determined to be better stabilizers for this use (Towler, 1984).

Gaonkar (1995) discussed the effects of stabilizer addition on drinkable yogurt. The initial increase, followed by the subsequent decrease in viscosity is shown as increasing levels of stabilizer are added. At the minimum viscosity, sedimentation is apparent (Gaonkar, 1995). The initial increase in viscosity results from the interaction between the proteins, which are positively charged, and the polysaccharides, which are negatively charged. Adding more stabilizer increases the charges on the particles, thereby increasing repulsion. This causes the decrease in viscosity and the increased sedimentation (Gaonkar, 1995). However, increasing the stabilizer content further increases the viscosity again and sedimentation is inhibited (Gaonkar, 1995). This effect is attributed to a decrease in the protein-polysaccharide interaction, allowing the stabilizer to exert an influence on the overall viscosity of the drinkable yogurt (Gaonkar, 1995).

Shukla *et al.* (1988) and Shukla and Jain (1991) studied the effects of gelatin, CMC, pectin, and other stabilizers on the organoleptic quality and the amount of whey

separation in yogurt made from buffalo milk. The use of 0.1 – 0.3% gelatin was determined to improve the appearance, body, texture, and flavor of the yogurts. Similarly, pectin (0.2-0.3%) improved these quality attributes and reduced whey separation. CMC, however, negatively impacted the quality of the yogurt and these samples were deemed unacceptable in sensory analysis (Shukla *et al.*, 1988; Shukla and Jain, 1991). The authors recommended that the usage of CMC not exceed 0.1% (Shukla *et al.*, 1988; Shukla and Jain, 1991). Another study by Shukla *et al.* (1987) researched the effects of these stabilizers on diacetyl and other flavor compounds associated with yogurt. The use of stabilizers was determined to decrease diactyl and volatile fatty acids in flavor compounds. This decrease was attributed to the stabilizers possible affecting the growth of *L. bulgaricus* (Shukla *et al.*, 1987). Jogdand *et al.* (1991) conducted similar work on *dahi*, a fermented milk product of India. The incorporation of gelatin and sodium alginate resulted in a decrease in titratable acidity, as well as total volatile acidity. Gelatin at 0.1% was determined to be sufficient to eliminate whey separation in *dahi* (Jogdand *et al.*, 1991).

Jawalekar *et al.* (1993) also examined the use of gelatin and other stabilizers related to yogurt rheology and sensory quality, as well as whey separation. The addition of gelatin to yogurt made with either cow or buffalo milk demonstrated an improvement in body, texture, viscosity, and curd tension. Whey separation was also reduced, likely due to the stabilizer binding free water in the yogurt (Jawalekar *et al.*, 1993). In addtition to gelatin, CMC, and pectin, numerous other stabilizers, such as starches, agar, locus bean gum, alginates, and guar gum have been studied for their use in yogurt (Tamime and Robinson,

1999). The properties, functionality, and quantity of these ingredients may all affect the mouthfeel and acceptance of a yogurt product.

Flavor and Sensory Acceptance

Yogurt can vary greatly in ingredient composition, so determining consumer preferences for these products is necessary for commercial success. Therefore, high importance is placed on flavor and texture of yogurt products. The characteristic yogurt flavor is attributed to acetaldehyde (Ott *et al.* 2000; Schmidt *et al.*, 1989; Vedamuthu, 1991; Wilkins *et al.*, 1986). Threonine is converted by the enzyme threonine aldolase to produce acetaldehyde, which imparts yogurt with a "green" or "green apple" flavor (assessed in plain yogurt) (Bodyfelt, 1988; Schmidt *et al.*, 1989; Wilkins *et al.*, 1986). According to Vedamuthu (1991), the optimum range for acetaldehyde in yogurt is 10-15 ppm. Other compounds have been shown to have impacts on the flavor and aroma of yogurt as well. For example, the diketones 2,3-butanedione (diacetyl) and 2,3-pentanedione are also contributors (Alonso and Fraga, 2001; Laye *et al.*, 1993; Ott *et al.*, 1997; Ott *et al.*, 2000).

Several studies have been conducted to evaluate the effects of the various ingredients and attributes on the sensory perception and preference of yogurt. Ott *et al.* (1997) determined that the acidity of yogurt plays a major role in yogurt flavor and is perhaps more important than the concentration of acetaldehyde, diacetyl, or 2,3-pentanedione. Added ingredients, such as stabilizers, may also influence flavor. Despite the possible benefits to yogurt quality, certain stabilizers may decrease the diacetyl content of yogurt (Jogdand *et al.*, 1991; Shukla *et al.*, 1987). This decrease may result from the stabilizers affecting the growth or interfering with the metabolic pathways of the starter cultures (Jogdand *et al.*, 1991; Shukla *et al.*, 1987). Fat content, which imparts both flavor and

mouthfeel, may be a contributor to flavor perception as well. Brauss *et al.* (1999) determined that low-fat yogurt released volatiles quicker and more intensely than the highest fat samples. The higher fat samples were also determined to have the highest viscosity.

As children are a substantial market population for yogurt, producers have developed products specifically designed to appeal to this group. These tend to be sweet, colorful, thick, and smooth (Gorski, 1994, Gorski, 1996; Brennan *et al.*, 2002). Research by Brennan *et al.* (2002) evaluated the responses to thickness and color on flavor intensity as perceived by 11-14-year-old panelists. Thickness did not influence the intensity of the flavor, but, along with flavor, was determined to affect preferences. Another study on flavor, color, and thickness preferences of children (ages 8-11) suggested that a yogurt drink could be designed to appeal to that age group (Papalia *et al.*, 1997).

Mouthfeel is also an important sensory property of yogurt products. According to Lawless and Heyman (1999), mouthfeel is defined as "a category of sensations occurring in the oral cavity, related to the oral tissues and their perceived condition (e.g. drying, coating)." These authors distinguish this property from texture, in which the sensations perceived are related to the actual food. Examples of this are firmness and hardness (Lawless and Heyman, 1999). For dairy products, chalkiness is a mouthfeel attribute that may be considered a defect (Wszolek *et al.*, 2001). In a comparison of yogurt made with added soy protein, Drake *et al.* (2000) described chalkiness as "the amount of fine chalk-like particles perceived when the yogurt is in the mouth." Research by Wszolek *et al.* (2001) on kefir, a fermented dairy beverage somewhat similar to drinkable yogurt, suggested that chalkiness and other mouthfeel properties may be influenced more by the

type of milk used, rather than effects of the starter cultures. Another factor that may contribute to a chalky mouthfeel is the choice of stabilizer used. The National Starch and Chemical Company emphasizes that hydrocolloid selection is crucial, as a particular stabilizer may either reduce or impart chalkiness, depending on the use and other properties (Maiolino, 2002). Certain conditions may also influence a chalky mouthfeel. Unstable casein, temperature extremes during processing, very rapid acid development, etc., have all been identified in yogurt as sources for a grainy mouthfeel, a similar defect to chalkiness (Bodyfelt, 1988).

Mouthfeel, flavor, sweetness, sourness, and the balance between these factors have been shown to affect the overall preference for yogurt (Barnes *et al.*, 1991). These characteristics, and many others, are important attributes for the acceptance of a drinkable yogurt product. Mouthfeel defects, such as chalkiness, may be affected by processing conditions, starter cultures, and stabilizer selection (Maiolino, 2002; Wszolek *et al.*, 2001). The objective of this research is to determine the effects of gelatin, CMC, and HMP on the mouthfeel of a drinkable yogurt, particularly related to chalkiness. Two different heat treatments will be applied to represent varying processing conditions and both ropy and nonropy starter cultures will be used.

CHAPTER 3
MATERIALS AND METHODS

Starter Cultures:

Ropy (Fargo 414) and a non-ropy (Fargo 404) starter cultures, provided by Quest International (Rochester, MN), were used for the production of yogurt. Cultures were aseptically transferred to sterile 10% nonfat milk and were incubated for 6 to 8 hours in a 37 °C water bath. Five subsequent subcultures were done by adding a 1.0% inoculum to sterile milk and incubated at 37 °C for 6 to 8 hours. After growth, the cultures were stored at 5 °C until use.

Yogurt Production:

Figure 1 shows the process used for making the drinkable yogurt for this study. Raw milk was obtained from the University of Florida Dairy Research Unit in Hague, Florida and transported to the pilot plant at the University of Florida, Food Science and Human Nutrition Department, Gainesville, Florida. The milk was warmed to 30-35 °C before being run through a separator (Clair Warenhandels GmbH Electric Cream Separator model FJ60AP, Treibach, Austria). Milk, standardized to 2% fat using appropriate amounts of cream and skim milk, was subdivided to test for the three stabilizers, gelatin (Type B Beef Skin Gelatin, 250/40 Mesh, Leiner Davis Gelatin, Jericho, NY), carboxymethylcellulose (CMC) (TICALOSE 2500 Pre-hydrated, TIC Gums, Inc., Belcamp, MD) and high methoxy pectin (HMP) (Pectin HM Rapid Set, TIC Gums, Inc. Belcamp, MD), two pasteurization regimes (68.3 °C

and 85 °C for 30 minutes) and for either strain of bacteria. The stabilizers used in this study were selected after noting their use in some drinkable yogurts available on the market, while the levels of stabilizers used were based upon preliminary studies in the laboratory. Drinkable yogurts were made with varying amounts of the stabilizers based upon recommendations by representatives from the gum suppliers and selected laboratory personnel evaluated these samples. The amount of stabilizer to be used in the investigation was determined by a general agreement on the thickness and acceptability of the yogurt. Pasteurization temperatures were chosen to represent a lower and higher range typical for the heat treatment of dairy products. The milk samples were warmed in a water bath to approximately 35-38 °C before the stabilizers were added (0.5% gelatin, 0.2% CMC, 0.1% HMP) under constant rapid stirring. Once the stabilizers had been thoroughly incorporated, the milk was pasteurized at the appropriate treatment temperature for 30 minutes.

The pasteurized milk was removed from the water bath and homogenized in a Gaulin Two Stage Laboratory Homogenizer, (Manton-Gaulin Manufacturing Co., Inc., Everett Massachusetts, model 15M-BTBA-10-08-03). The homogenization pressures were 2500 psi stage 1 and 500 psi stage 2. After homogenization, the milk was immediately cooled to 45 °C, inoculated with 2% yogurt culture of either the ropy or non-ropy strains and was allowed to incubate in a 45°C water bath until a pH of 4.0-4.3 was reached. The yogurt was refrigerated at 4 °C for 24 hours before the flavorings (strawberry puree, sugar, and corn syrup) were added.

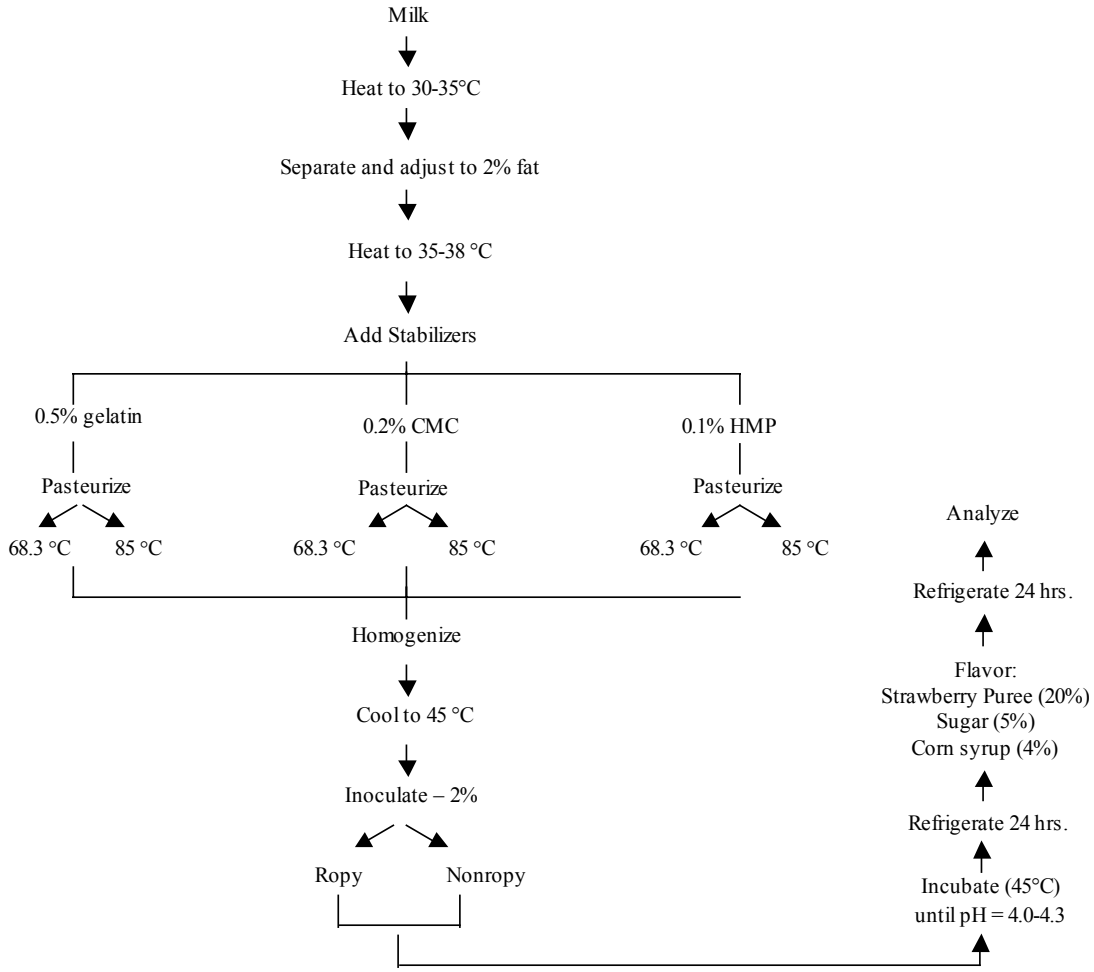


Figure 1 Processing Diagram for Drinkable Yogurt Production

Strawberries obtained from a local supermarket were pureed using a Waring Blendor and strained through a two-screened sieve (inner screen: 1mm x 1mm mesh, outer screen 3mm x 3 mm mesh) to remove the majority of the seeds. Sucrose, light corn syrup, and strawberry puree were added to the yogurt in the amounts of 4%, 5%, and 20%, respectively. The flavorings were blended gently with the yogurt for 20 seconds on the slowest setting of a Kitchen-Aid mixer (model K5-A, The Hobart Manufacturing Company, Troy, Ohio). The yogurt was refrigerated for at least 24 hours before analyses. The treatments made for this study are listed in Table 1.

Table 1 Drinkable Yogurt Treatments

Pasteurization Temperature (°C)[1]	Stabilizer Added[2]	Starter Culture[3]	Designation
68.3	Gelatin	Nonropy	68.3GelNR
85	Gelatin	Nonropy	85GelNR
68.3	CMC	Nonropy	68.3CMCNR
85	CMC	Nonropy	85CMCNR
68.3	HMP	Nonropy	68.3HMPNR
85	HMP	Nonropy	85HMPNR
68.3	Gelatin	Ropy	68.3GelR
85	Gelatin	Ropy	85GelR
68.3	CMC	Ropy	68.3CMCR
85	CMC	Ropy	85CMCR
68.3	HMP	Ropy	68.3HMPR
85	HMP	Ropy	85HMPR

1 30 minute heating time, 2 Gelatin – 0.5%, CMC – 0.2%, HMP – 0.1%, 3 Nonropy = Fargo 404, Ropy = Fargo 414; Inoculation at 2% (w/v)

Sensory Analysis

Fifteen volunteers (7 females, 8 males) were trained in ten sessions to evaluate attributes related to the flavor and mouthfeel of the drinkable yogurt samples. During the first training session, important sensory characteristics of drinkable yogurt were identified. The descriptors chosen were: viscosity, chalkiness, mouth coating, yogurt flavor intensity, strawberry flavor intensity, sourness, off-flavor, and overall acceptability. Reference samples, provided during the next three training sessions as examples of these attributes, were as follows:

Viscosity – commercial cup yogurt (Dannon, Minster, OH), commercial yogurt packaged in tubes (Yoplait Exprèsse, General Mills, Inc., Minneapolis, MN), commercial drinkable yogurts (Dannon Danimals, Minster, OH, and Yonique, Cacique, Inc., City of Industry, CA). These references served as examples of high, medium, and low viscosity ranges, respectively.

Chalkiness – defined as both a taste and a feeling of chalky particles on the tongue and behind the teeth during swallowing; Antacid tablets (Tums Ultra assorted mixed berry flavor) ground into a powder (150 g total) using a Waring Blendor (model 31BL92, Waring Products Division, Dynamics Corporation of America, New Hartford, CT) and mixed with 400 ml distilled water.

Mouth coating – heavy whipping cream (Publix Supermarkets, Lakeland, FL). It should be noted that this did not necessarily represent the actual mouth coating of a drinkable yogurt product, but was as an example of the general term to ensure all panelists would be evaluating the same characteristic.

Yogurt flavor intensity – Nonfat plain yogurt (Dannon, Minster, OH)

Strawberry flavor intensity – fresh strawberries, pureed

Sourness – Nonfat plain yogurt (Dannon, Minster, OH)

No reference was provided for off-flavor, as this term was used to encompass any flavor that might be present but could not be associated with yogurt or strawberries.

During the next four training sessions, panelists were trained by rating drinkable yogurt samples manufactured as described in Figure 1. The various attributes were presented on paper ballots and panelists were asked to rate the samples on 15 cm line scales for each attribute. Results were compared and discussed as a group to ensure that the panelists were scoring the attributes in the same general ranges on the scale. Panelists who did not evaluate the attributes correctly were given additional training.

Computerized sample ballots (Compusense Five, version 4.2, Guelph, Ontario, Canada) were used for practice during the last two training sessions. This was done

to familiarize the panelists with the exact ballots to be used for testing. An example of the ballot used for the sensory testing is depicted in Figure 2.

Sensory analysis was conducted on the experimental yogurt within four days of production. Drinkable yogurts made with the ropy starter cultures were evaluated on different days than those made with the nonropy cultures. All sensory analyses were performed in individual booths with white lighting. Unsalted crackers and distilled water were provided for panelists to cleanse their palates before and during tasting. At each tasting session, panelists were presented with a tray of six samples, each coded with a different, random, three-digit number. The samples were distributed so all treatments were presented in the each position on the tray the same number of times. Approximately 60 ml of each drinkable yogurt treatment were poured into 118 ml opaque, plastic cups. Panelists were asked to evaluate the six treatments, with instructions to take a short break between the third and fourth samples.

Qualitative Descriptive Analyses were preformed using Compusense Five (version 4.2) software to measure the attributes selected during the training sessions. Overall Viscosity, Chalkiness, Mouthcoating, Yogurt Flavor, Strawberry Flavor, Sourness, Off-flavor, and Overall Acceptability were rated on a 15 cm line scale. Anchors for all attributes except viscosity were listed as 'None' (0 cm) to 'High' (15 mm). For measuring Overall Viscosity, a score of 0 cm indicated a 'Thin' sample, while 15 cm was considered 'Thick'. Replications were conducted the following day. In total, two batches of yogurt for each type of starter culture used, with subsequent replications, were evaluated.

Whey Separation

Whey separation was measured using gravity separation. One hundred milliliters of each sample were poured into glass graduated and placed on an undisturbed shelf in a 2.8 °C cooler. Separation of the serum fluid from the gel matrix was visually measured after storage under refrigeration for 5 days. All samples were evaluated in triplicate.

pH and Titratable Acidity

Yogurt pH was measured using a Fisher Scientific Accumet pH meter with a combination pH/ATC Accumet electrode. Titratable acidity (TA) was measured by titration using 0.1N sodium hydroxide, as listed for yogurt in the Standard Methods for the Examination of Dairy Products (Richardson, 1985).

Rheology

Viscosity was measured using an AR2000 Rheometer (TA Instruments, Inc, New Castle, DE) using a conical concentric cylinder (15 mm stator inner radius, 14 mm rotor outer radius, 42 mm cylinder immersed height, continuous ramp step). Samples were tested at 5 °C. Data was recorded and analyzed using the Rheology Advantage Data Analysis software (version 3.0.24).

Protein Analysis

Soluble protein was measured using the Lowry Method (Lowry *et al.*, 1951). One milliliter of each drinkable yogurt sample was diluted in 100 ml distilled water and allowed to sit. Bovine serum albumin was used as a standard. Dilutions of 0.1, 0.2, 0.3, 0.5, 0.7, and 1.0 ml of drinkable yogurt samples were tested. Three milliliters of each prepared sample dilution were placed in individual wells of a microplate and the absorbance was read at 650 nm in a spectrophotometer (Coulter Microplate Reader).

Statistical Analyses

Sensory attributes, pH, titratable acidity, and whey separation data were analyzed using ANOVA (SAS,Version 8.2, SAS Institute, Cary, NC) for a complete 3 x 2 x 2 factorial design with stabilizer, heating temperature, and starter culture as the factors. The levels were gelatin, CMC, and HMP for stabilizer, 155 °F and 185 °F for heating temperature, and ropy and nonropy for the starter cultures. Interactions and simple effects of the significant attributes were evaluated using Students t-tests.

Soluble protein data were analyzed using simple linear regression and pairwise comparisons. The data was further analyzed by t-tests.

Drinkable Yogurt

Please taste the samples in the order presented. Indicate your response for each attribute by making a mark (labeled with the cup number) on the appropriate line. Crackers and water will be provided to cleanse your palate between samples.

Overall Viscosity:

Thin — Thick

Chalkiness:

None — High

Mouthcoating:

None — High

Yogurt Flavor Intensity:

None — High

Strawberry Flavor Intensity:

None — High

Sourness:

None — High

Off-flavor:

None — High

Overall Acceptability:

None — High

Comments:

Figure 2 Sample Ballot for Sensory Analysis

CHAPTER 4
RESULTS AND DISCUSSION

Visual observation of the drinkable yogurts showed that the samples made with CMC were very different than those made with gelatin or HMP. While products made with the latter two stabilizers were thicker, light pink in color, and more homogeneous, the CMC samples were thin, dark pink, and had large amounts of aggregated protein. These chunks could not be incorporated into the yogurt by mixing, resulting in a very unpleasant consistency. Hence, the samples made with CMC were completely unacceptable as drinkable yogurt.

Sensory Analysis

Tables 2 and 3 list the means and F-values for all attributes evaluated during sensory analyses. The type of stabilizer used in drinkable yogurt production had a significant effect on the overall viscosities of the samples ($p<0.05$). The range of mean sensory viscosity scores was 1.3 to 12.4 cm. Yogurts made with both ropy and non-ropy cultures and gelatin were the thickest yogurts, followed by the yogurts made with HMP (Figure 3). The samples made with CMC were the thinnest, which agrees with Phillips and Williams (2000), who suggest that, although the CMC/casein complexes are stable upon heating, viscosity tends to be decreased. In comparison to the ropy samples, the viscosity was significantly ($p<0.05$) higher for drinkable yogurts made with nonropy cultures and HMP than those made ropy cultures and the same stabilizer. This is also seen for samples made with nonropy cultures and stabilized with CMC, but the difference in

cultures had no significant effect on the viscosity of drinkable yogurts made with gelatin (Table 2 and 3).

Table 2 Mean Scores of the Sensory Characteristics of Drinkable Yogurts

Treatment			Overall Viscosity	Chalkiness	Mouth Coating	Yogurt Flavor Intensity	Strawberry Flavor Intensity	Sourness	Off-Flavors	Overall Acceptability
Past.[2] Temp. (°C)	Stabilizer[3]	Culture[4]								
68.3	Gelatin	Nonropy	12.2	6.6	9.4	9.7	9.0	8.3	1.4	10.6
85.0	Gelatin	Nonropy	12.4	6.9	9.5	10.1	8.4	9.4	1.4	9.9
68.3	CMC	Nonropy	1.5	5.4	3.4	4.7	7.8	5.5	2.8	3.1
85.0	CMC	Nonropy	2.8	8.1	5.6	4.6	7.0	5.8	3.2	2.2
68.3	HMP	Nonropy	10.4	8.6	8.5	9.5	8.4	8.5	1.6	10.1
85.0	HMP	Nonropy	10.7	8.5	8.8	9.2	9.3	8.3	1.3	10.2
68.3	Gelatin	Ropy	12.0	6.6	9.0	9.4	8.9	7.0	1.5	11.1
85.0	Gelatin	Ropy	12.2	6.6	8.5	9.2	8.9	7.1	1.2	10.9
68.3	CMC	Ropy	1.4	6.0	2.9	4.6	6.8	4.9	3.9	2.1
85.0	CMC	Ropy	1.3	6.0	3.0	4.1	6.8	4.3	3.8	2.1
68.3	HMP	Ropy	8.4	7.8	6.4	9.0	9.2	8.0	1.4	9.9
85.0	HMP	Ropy	10.1	8.0	7.3	9.0	8.7	6.3	1.3	10.7

[1]Sensory Characteristics rated on a 15 cm line
[2]30 minute heating time
[3]Gelatin = 0.5%; CMC = 0.2%; HMP = 0.1%
[4]Nonropy = Fargo 404; Ropy = Fargo 414; Inoculation at 2% (w/v)

Table 3 F-values and Levels of Significance for Factors and Interactions with the Sensory Attributes

Variables	Sensory Characteristics							
	Viscosity	Chalkiness	Mouth Coating	Yogurt Flavor Intensity	Strawberry Flavor Intensity	Sourness	Off-flavors	Overall Acceptability
Culture	14.78*	0.53(ns)	8.30*	0.69(ns)	0.04(ns)	10.89*	0.39(ns)	0.47(ns)
Temperature	19.89**	6.00*	13.90*	1.61(ns)	1.06(ns)	2.84(ns)	0.20(ns)	0.73(ns)
Temp*Culture	0.01(ns)	8.08*	3.21(ns)	0.96(ns)	0.01(ns)	12.90*	1.04(ns)	5.25*
Stabilizer	2393.39**	4.79*	122.36**	199.45**	27.46**	30.69**	41.79**	317.48**
Stab*Culture	11.10*	0.42(ns)	1.86(ns)	0.79(ns)	2.13(ns)	0.65(ns)	1.87(ns)	2.14(ns)
Temp*Stab	1.66(ns)	4.24*	2.75*	1.23(ns)	0.51(ns)	5.68*	0.67(ns)	3.65*

* = significant at $p<0.05$ level
** = significant at $p<0.01$ level
(ns) = not significant

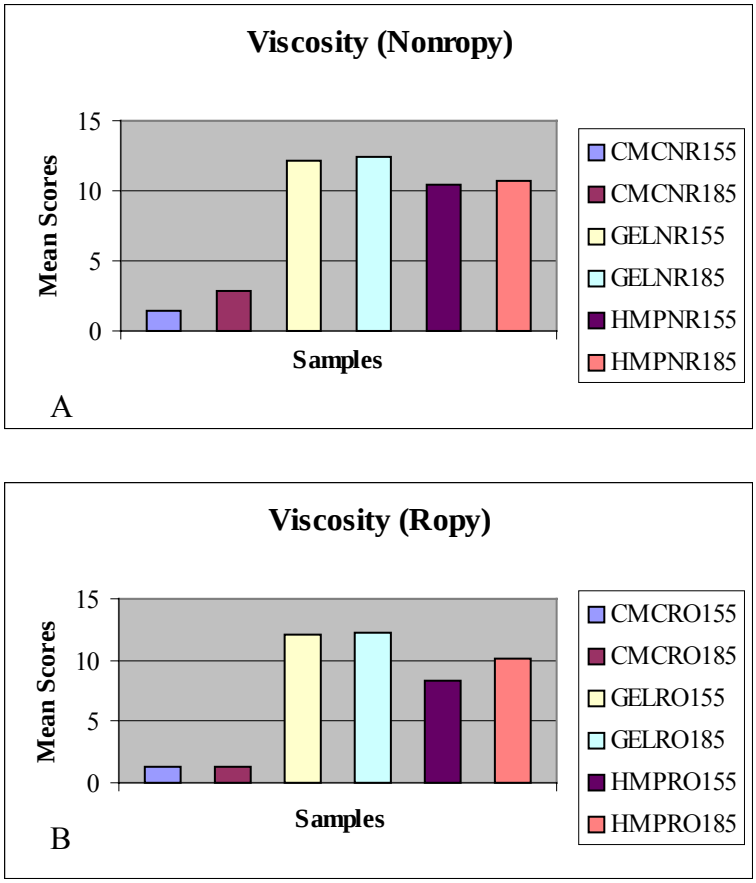


Figure 3 Mean Sensory Scores for Viscosity. A) Drinkable Yogurts Made With Nonropy Cultures (Fargo 404; Inoculation at 2% w/v); B) Drinkable Yogurts Made With Ropy Cultures (Fargo 414; Inoculation at 2% w/v).

The type of stabilizer also influenced perceived yogurt chalkiness, as seen in Tables 2 and 3. The average of mean chalkiness scores ranged from 5.4 to 8.6 cm on a 15 cm line scale. For yogurt that had been pasteurized at 68.3 °C, HMP was determined to be significantly ($p<0.05$) chalkier than products made with the other two hydrocolloids at that pasteurization treatment. Yogurts made with gelatin were the second highest in chalky character, while the CMC samples were the least chalky. Like those heated at 68.3 °C, the samples made with HMP and heated to 85 °C were the most significantly ($p<0.05$) chalky. However, chalkiness was not affected by the presence of gelatin or CMC for this heat treatment. An interaction between the pasteurization temperature and

type of culture used was also analyzed. The type of culture made no significant difference on chalkiness for the products heated to 68.3 °C, but drinkable yogurts made from nonropy cultures and heated to 85 °C tested higher in chalkiness than at 68.3 °C (Figure 4).

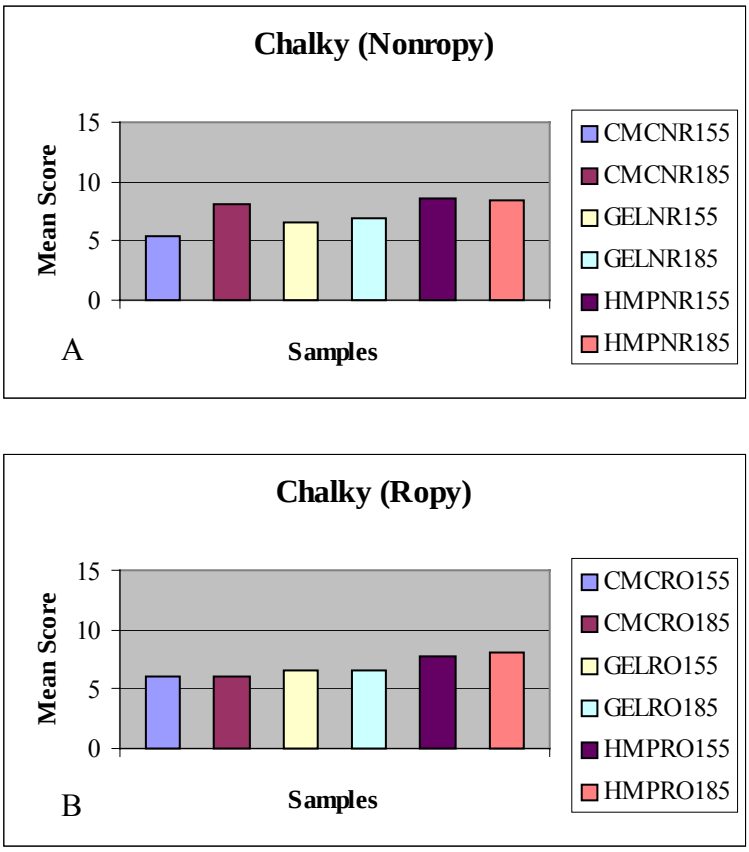
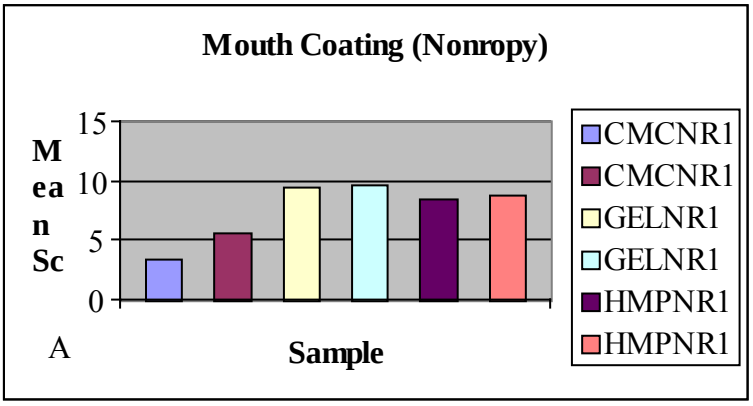


Figure 4 Mean Sensory Scores for Chalkiness. A) Drinkable Yogurts Made With Nonropy Cultures (Fargo 404; Inoculation at 2% w/v); B) Drinkable Yogurts Made With Ropy Cultures (Fargo 414; Inoculation at 2% w/v).

Mouth coating was significantly affected by culture selection, heating temperature, and type of stabilizer used (Tables 2 and 3, Figure 5). The mean mouth coating scores ranged from 2.9 to 9.5 cm on a 15 cm line scale. Mouth coating was less for yogurts pasteurized at the lower temperature, with the exception of the samples made with gelatin. The samples made with gelatin also showed no differences between samples

made with ropy and nonropy starter cultures. However, mean scores for mouth coating were lower for drinkable yogurts made with ropy cultures and stabilized with either HMP or CMC than comparable samples made with nonropy cultures. In terms of the stabilizers, the samples made with gelatin were the most mouth coating, followed by those made with HMP.



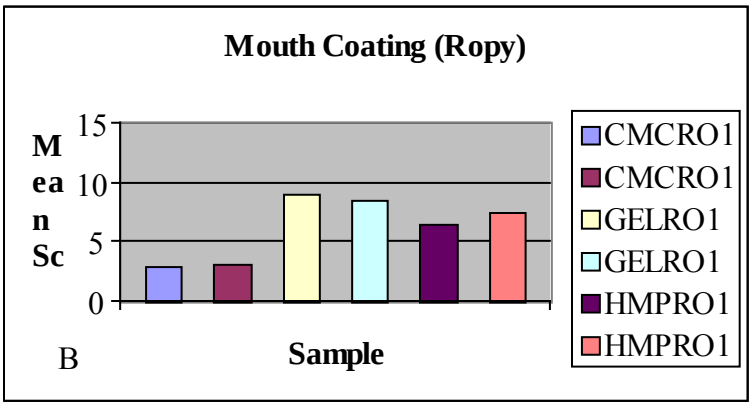


Figure 5 Mean Sensory Scores for Mouth Coating. A) Drinkable Yogurts Made With Nonropy Cultures (Fargo 404; Inoculation at 2% w/v); B) Drinkable Yogurts Made With Ropy Cultures (Fargo 414; Inoculation at 2% w/v).

The type of stabilizer used also affected the yogurt flavor intensity (Tables 2 and 3). The mean scores for flavor intensity ranged from 4.1 to 10.1 cm on a 15 cm line scale. Figure 6 depicts the means for the yogurt flavor intensity attribute. As shown, no differences were found between the yogurt flavor intensity of drinkable yogurts made with gelatin and HMP. These two products, however, had more intense yogurt flavor

than the samples made with CMC. Similarly, the type of stabilizer used also affected the strawberry flavor intensity, as can be seen in Tables 2 and 3. Like yogurt flavor intensity, yogurts made with gelatin and HMP were higher in strawberry flavor intensity (mean range: 6.8 to 9.3 cm on a 15 cm line scale) than the products made with CMC, but no differences were determined between yogurts made with the former two stabilizers (Figure 7).

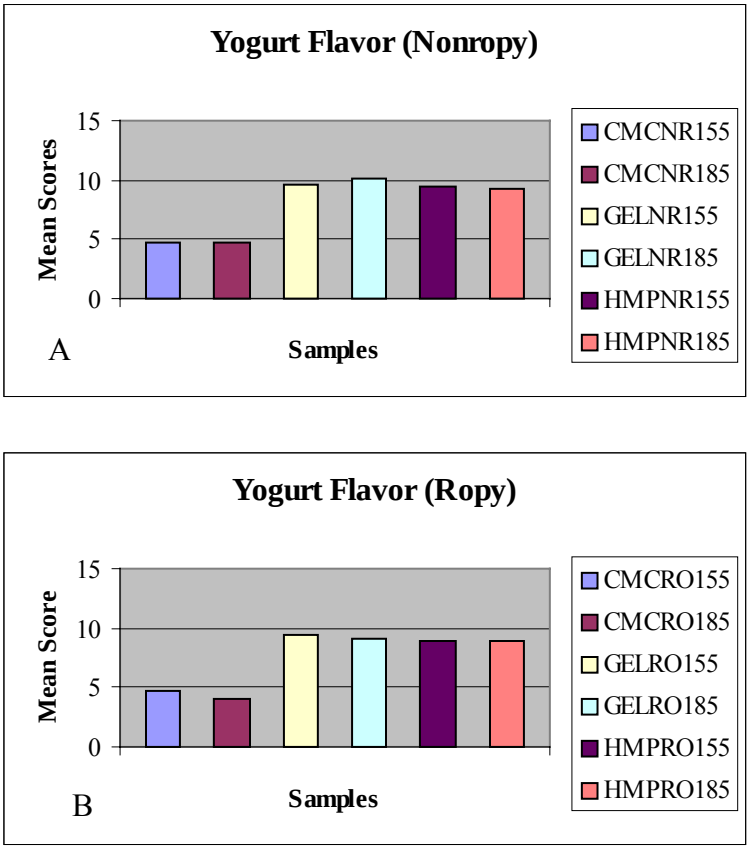


Figure 6 Mean Sensory Scores for Yogurt Flavor Intensity. A) Drinkable Yogurts Made With Nonropy Cultures (Fargo 404; Inoculation at 2% w/v); B) Drinkable Yogurts Made With Ropy Cultures (Fargo 414; Inoculation at 2% w/v).

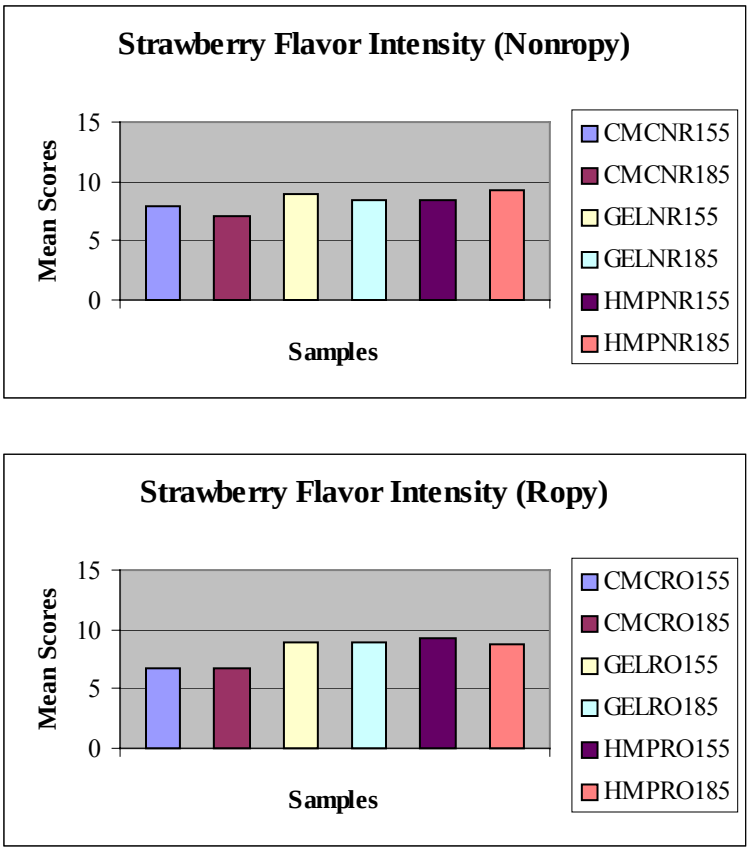


Figure 7 Mean Sensory Scores for Strawberry Flavor Intensity. A) Drinkable Yogurts Made With Nonropy Cultures (Fargo 404; Inoculation at 2% w/v); B) Drinkable Yogurts Made With Ropy Cultures (Fargo 414; Inoculation at 2% w/v).

Heating temperatures, stabilizers, and starter cultures all play a role in the sour perception by panelists (Tables 2 and 3). The mean scores for sourness ranged from 4.3 to 9.4 cm on a 15 cm line scale. Yogurts fermented by the ropy cultures were perceived as being significantly ($p<0.05$) less sour than those made from nonropy cultures at both 68.3 °C and 85 °C. The sourness rating was also significantly ($p<0.05$) lower for drinkable yogurts made with ropy cultures and heated to 85 °C as compared to those heated to 68.3 °C. This effect was not observed in the yogurts made with nonropy cultures, as demonstrated in Figure 8. Yogurts that had been pasteurized at 68.3 °C and made with gelatin or HMP were significantly more sour than those made with CMC at the same temperature. Despite this, samples pasteurized at 68.3 °C with gelatin as the

stabilizer and those pasteurized at 85 °C with HMP, as the stabilizer displayed no differences in sourness rating. However, for yogurts heated at 85°C, those made with gelatin were perceived as being more sour than those made with HMP, while drinkable yogurts made with CMC were the least sour.

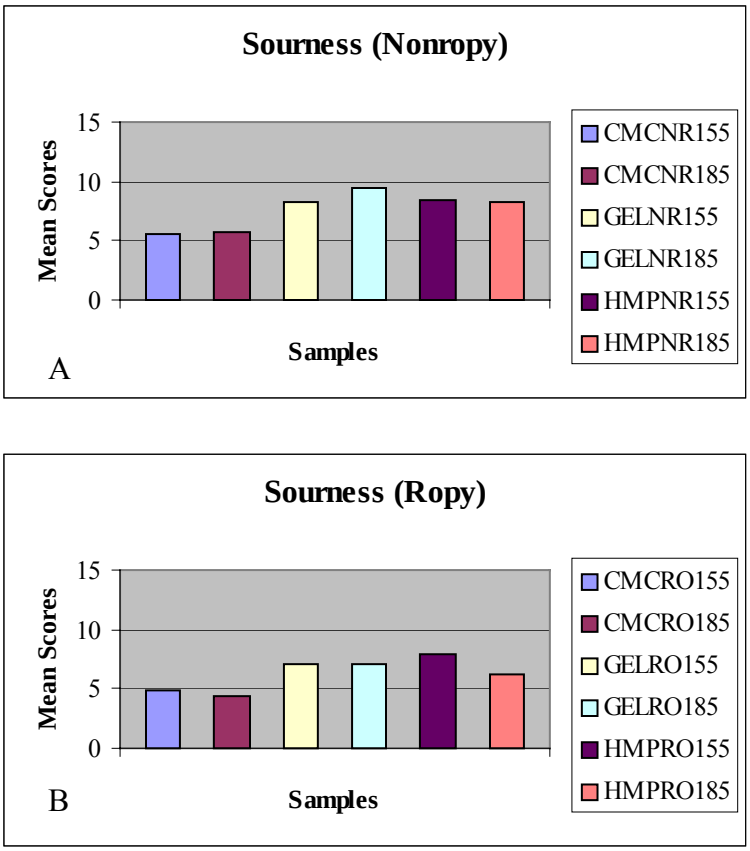


Figure 8 Mean Sensory Scores for Sourness. A) Drinkable Yogurts Made With Nonropy Cultures (Fargo 404; Inoculation at 2% w/v); B) Drinkable Yogurts Made With Ropy Cultures (Fargo 414; Inoculation at 2% w/v).

Off-flavors were found in the samples containing CMC (Tables 2 and 3). Despite being measured on a 15 cm line scale, this information may not be completely representative of the samples. Since CMC was so different than the samples made with the other two stabilizers, this would account for a higher off-flavor scoring for this sample. As can be seen by the data in Tables 2 and 3, off-flavors were minimal in samples made with gelatin and HMP. Another possible source of any off-flavors might

be attributed to the strawberry puree used for flavoring the drinks. Although fresh strawberries were used for all tests, changes in the flavor and aroma of the strawberry puree were noted after a few days. The "fresh" quality of the puree seemed to be diminished after a short time and a more jam-like flavor and aroma was apparent. Though yogurt samples were tested within four days of production, these altered notes may have contributed to an increased panelist response to off-flavor.

No differences in overall acceptability (mean score range: 2.1 to 10.9 cm on a 15 cm line scale) were determined between drinkable yogurts pasteurized at 68.3 °C and stabilized with gelatin and 68.3 °C stabilized with HMP as compared to their respective counterparts heated at 85 °C. However, as seen in Tables 2 and 3, stabilizers did play a role in overall acceptability when compared against each other. The mean scores reported for overall acceptability is depicted in Figure 9. Among those heated at 85 °C, drinkable yogurts made with gelatin and HMP were not different ($p<0.05$) from each other. Drinkable yogurt samples with gelatin and heated at 68.3 °C were found to be more acceptable than those made with HMP and heated to the same temperature. The use of ropy cultures for samples heated to 85 °C produced slightly more acceptable products than drinkable yogurts made with nonropy cultures at the same temperature. However, no differences were determined ($p>0.05$) for yogurts heated to 68.3 °C using either type of starter culture.

The samples made with CMC were found to be the least acceptable. This result was expected, as CMC was added prior to fermentation, resulting in protein agglomeration and a large degree of whey separation and other defects. Thus, the CMC yogurt samples were very thin with large particles and chunks. Samples made with CMC were

considered by the panelists unpleasant to drink and would not be commercially acceptable. Hence, these products cannot be compared to the gelatin- and HMP-stabilized yogurt products. Imeson (1997) suggests adding CMC after fermentation to prevent these defects, which was not done in this study.

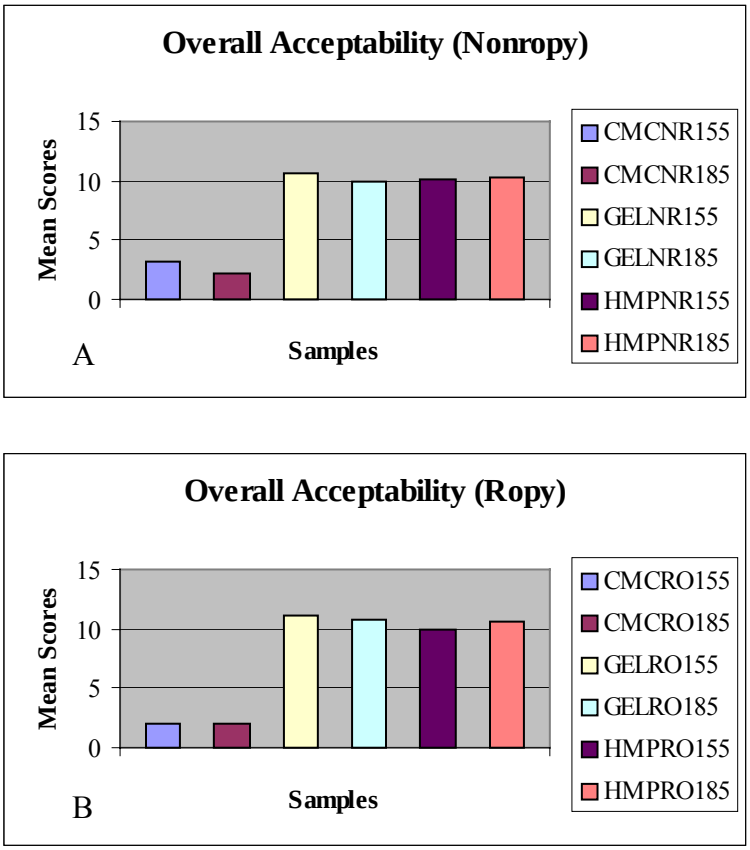


Figure 9 Mean Sensory Scores for Overall Acceptability. A) Drinkable Yogurts Made With Nonropy Cultures (Fargo 404; Inoculation at 2% w/v); B) Drinkable Yogurts Made With Ropy Cultures (Fargo 414; Inoculation at 2% w/v).

pH and Titratable Acidity

The pH for samples heated to 68.3 °C and 85 °C made with ropy starter cultures were significantly higher than the nonropy cultures processed under the same conditions. Drinkable yogurt heated to 85 °C and fermented with ropy cultures had significantly higher pH values than samples heated to 68.3 °C. No difference was found among the

nonropy cultures. Table 4 lists the means for the pH measurements, while Table 5 gives the F-values for the samples.

Titratable acidity (TA) values, expressed as % lactic acid, are listed in Table 4 and Table 5. TA was significantly higher ($p<0.05$) for samples stabilized with gelatin and HMP and fermented with nonropy starters than the comparable products made with ropy cultures. In the nonropy samples, no difference was determined in the TA for yogurts made with gelatin and HMP, but these products were higher in TA than the CMC samples. In the ropy samples, TA values were significantly higher for products stabilized with HMP than CMC, but there were no differences between those with gelatin and CMC and gelatin and HMP. The low values for TA agree with results from Jogdand *et al.* (1991) on the study of *dahi*, a fermented milk product from India. The presence of

Table 4 Mean Scores for the pH, Titratable Acidity, and Whey Separation Data

Treatment			pH	% lactic acid	% whey separation
Past. Temp. (°C)	Stabilizer	Culture			
68.3	Gelatin	Nonropy	3.82	1.088	0.0
85.0	Gelatin	Nonropy	3.80	1.124	0.0
68.3	CMC	Nonropy	3.92	0.803	24.0
85.0	CMC	Nonropy	3.99	0.782	10.3
68.3	HMP	Nonropy	3.85	1.056	0.0
85.0	HMP	Nonropy	3.79	1.080	0.0
68.3	Gelatin	Ropy	3.95	0.912	0.0
85.0	Gelatin	Ropy	3.99	0.873	0.0
68.3	CMC	Ropy	3.99	0.905	22.0
85.0	CMC	Ropy	4.07	0.767	8.0
68.3	HMP	Ropy	3.89	0.954	0.0
85.0	HMP	Ropy	3.96	0.885	0.0

Table 5 F-Values and Levels of Significance for Factors and Interactions with the pH, Titratable Acidity, and Whey Separation Measurements

Variables	pH	% lactic acid	% whey separation
Temperature	5.55*	2.33$^{(ns)}$	8.67*
Stabilizer	33.26*	29.02*	68.72*
Culture	73.10*	21.98*	0.04$^{(ns)}$
Temperature*Culture	7.26*	4.41$^{(ns)}$	0.60$^{(ns)}$
Stabilizer*Culture	3.24$^{(ns)}$	11.61*	0.04$^{(ns)}$
Temperature *Stabilizer	3.09	1.06$^{(ns)}$	8.19*

* = significant at $p<0.05$ level, $^{(ns)}$ = not significant

stabilizers lowered the TA of the products. This may result from interference in either the metabolism of the starter cultures or the growth, causing a lower production of acids. The addition of the strawberry puree in this experiment may also have contributed to these TA values.

Whey Separation

Five days after flavoring the yogurt, separation was measured (means displayed in Figure 10). In that time, none of the gelatin or HMP samples exhibited any separation. However, all of the CMC samples separated, usually starting on the first day. The average percent of separation for these products are listed in Table 4. The amount of settling was significantly more for the CMC treatment 68.3 °C than the treatment at 85 °C. Both treatments heated to 68.3 °C demonstrated greater whey separation, which is indicative of instability in the gel structure (Table 5). The lack of whey separation for the drinkable yogurt samples made with gelatin and HMP may indicate that there was a sufficient amount of negatively-charged hydrocolloid to provide repulsion on the positively-charged protein molecules of the yogurt, thereby stabilizing the matrix (Gaonkar, 1995). CMC, however, was added to the milk at a pH above the isoelectric

point of casein. As a result, the CMC formed complexes with the milk proteins that precipitated out of solution. Hence, CMC used in this manner is unacceptable to prevent whey separation. This is supported by the results of Shukla and Jain (1991), who found similar results in yogurt made from buffalo milk. As in this experiment, Shukla and Jain (1991) also found pectin and gelatin to be useful stabilizers to prevent whey separation. Jawalekar *et al.* (1993) and several other researchers determined that gelatin reduces whey separation of yogurt.

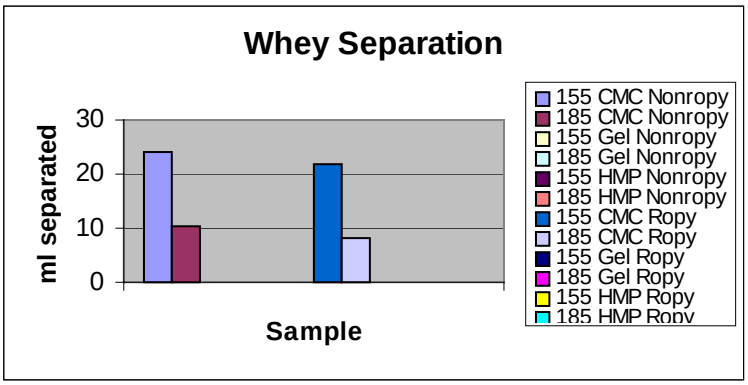


Figure 10 Mean Scores for Whey Separation for Drinkable Yogurts

Protein Analysis

Protein content was analyzed for all drinkable yogurts produced. Table 6 shows the mean protein contents for each of the samples. Mean scores ranged from 0.245 mg/ml to 0.858 mg/ml protein. In terms of ropy vs. nonropy samples, no significant differences were found. The exceptions to this were for the drinkable yogurts heated to 85 °C and stabilized with either CMC or HMP. In these, the protein contents were higher for the nonropy yogurts than ropy. Temperature effects were also apparent. Protein content was lower for gelatin-stabilized and CMC-stabilized yogurts made with the ropy cultures and pasteurized at 85 °C, as compared to their 68.3 °C counterparts. No differences were

Table 6 Mean Scores for Protein Content for All Drinkable Yogurt Samples

Treatment			Protein content (mg/ml)[3]
Past. Temp. (oC)	Stabilizer[1]	Culture[2]	
68.3	Gelatin	Nonropy	0.811
68.3	CMC	Nonropy	0.259
68.3	HMP	Nonropy	0.565
85.0	Gelatin	Nonropy	0.490
85.0	CMC	Nonropy	0.245
85.0	HMP	Nonropy	0.538
68.3	Gelatin	Ropy	0.580
68.3	CMC	Ropy	0.466
68.3	HMP	Ropy	0.550
85.0	Gelatin	Ropy	0.858
85.0	CMC	Ropy	0.278
85.0	HMP	Ropy	0.514

[1]Gelatin = 0.5%; CMC = 0.2%; HMP = 0.1%
[2]Nonropy = Fargo 404; Ropy = Fargo 414
[3]Determined by Lowry Method

determined for samples made with HMP and ropy cultures at either temperature, or any nonropy yogurts samples. Stabilizer interactions were also analyzed for the measurements of protein content. For the nonropy samples, samples stabilized with CMC were significantly lower than those stabilized with HMP. Yogurt products made with gelatin or HMP and ropy starter cultures were both higher in protein content than CMC samples made with the same cultures. There was no difference, however, in protein content of drinkable yogurts fermented with ropy cultures and containing gelatin or HMP.

Rheology

Figures 11 – 16 depict the shear stress vs. shear rate and viscosity vs. shear rate for all drinkable yogurt samples. The drinkable yogurt samples produced for this experiment exhibited pseudoplastic behavior (shear thinning). This non-Newtonian flow was

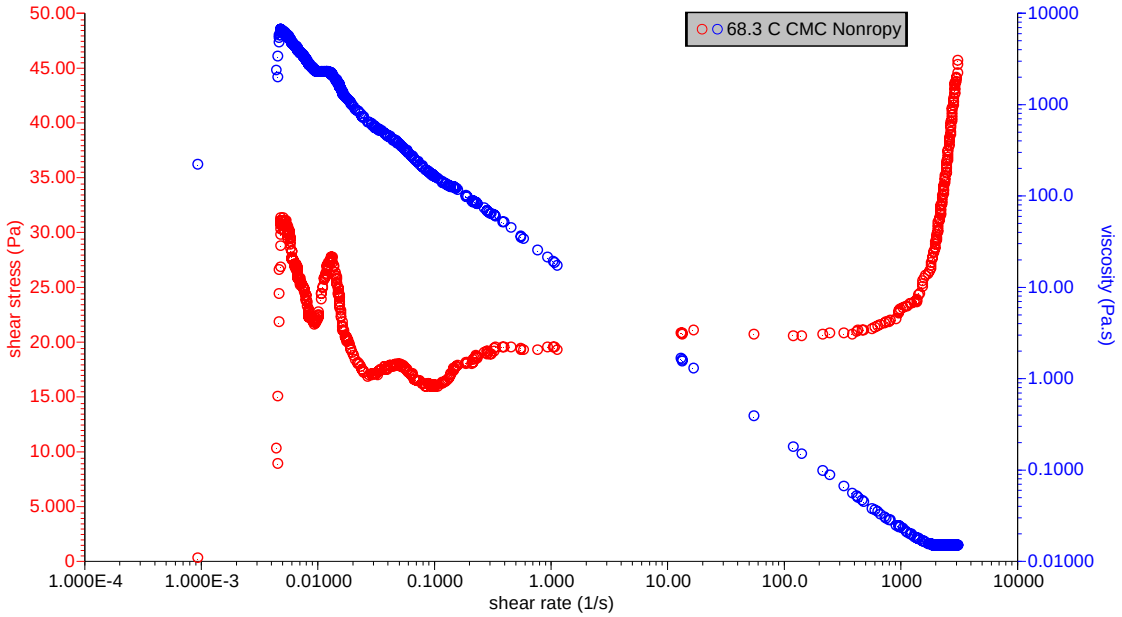
expected, as is commonly found in yogurts and related products (Keogh and O'Kennedy, 1998; Rawson and Marshall, 1997; Schmidt *et al.*, 1980; Schmidt and Smith, 1992). Hence, the Power Law Model was utilized for the rheology data. The equation for this model is: $\sigma = k * \gamma^n$, where σ = shear stress, k = consistency coefficient, γ = shear strain, and n = power law factor.

The consistency coefficients (k), which typically correlate to the viscosity, were significantly higher for samples made with gelatin and HMP fermented with ropy cultures compared to those made with CMC and ropy cultures. This agreed with visual observation that gelatin and HMP samples were noticeable thicker. No significant differences, however, were determined among nonropy samples. However, the value obtained for samples made with CMC may not be accurate. The large amount of aggregated protein most likely interfered with the rotational movement of the bob in the cup, resulting in a higher value. This is supported by the fact that CMC samples made with nonropy starter cultures were thinner than gelatin and HMP samples, though the measured k values do not represent this fact. For that matter, that the k values for the CMC samples made with nonropy cultures is significantly higher ($p<0.05$) than the CMC samples made with ropy cultures is likely not reliable as well. However, no differences were determined in the k values when comparing drinkable yogurts made with gelatin to those made with HMP, regardless of the starter culture used. These findings agree with those of Rawson and Marshall (1997) in that the ropy strains did not necessarily influence the viscosity. This suggests that the protein interactions in the yogurt matrix play a larger role in viscosity than the polysaccharide-protein interactions of the ropy bacteria (Rawson and Marshall, 1997). The heat treatment was shown to have an influence on the

k value. Drinkable yogurts pasteurized to 85 °C had significantly higher ($p<0.05$) k values than those pasteurized to 68.3 °C. The thickness of the samples made with gelatin agreed with previous research by Keogh and O'Kennedy (1998). In that study, gelatin increased the k and decreased n. Barak and Ramaswamy (1994) determined similar results for yogurts made with pectin and strawberry concentrate. However, these authors concluded that the strawberry concentrate had more of an effect on the consistency coefficient than the pectin. Pectin did increase the flow behavior index, though the strawberry concentrate decreased the value (Barak and Ramaswamy, 1994).

The power law factor for the drinkable yogurts produced ranged from 0.014 to 0.147 (Table 7). The yogurt made with CMC and ropy cultures demonstrated the least amount of shear thinning, which was expected since the protein in these samples aggregated and the yogurt matrix was not maintained. Therefore, the CMC samples were not representative of an acceptable drinkable yogurt, regardless of their small departure from pseudoplasticity. The gelatin and HMP samples both exhibited some shear thinning behavior, though no differences were determined between samples made with ropy and nonropy starter cultures. Heat treatment did not affect the power law factor.

Although strained to reduce the amount of seeds from the addition of the strawberry puree, some seeds remained in the samples. In some cases, these seeds may have altered the overall rheometer readings. Eliminating the seeds completely may yield more consistent results in future studies.



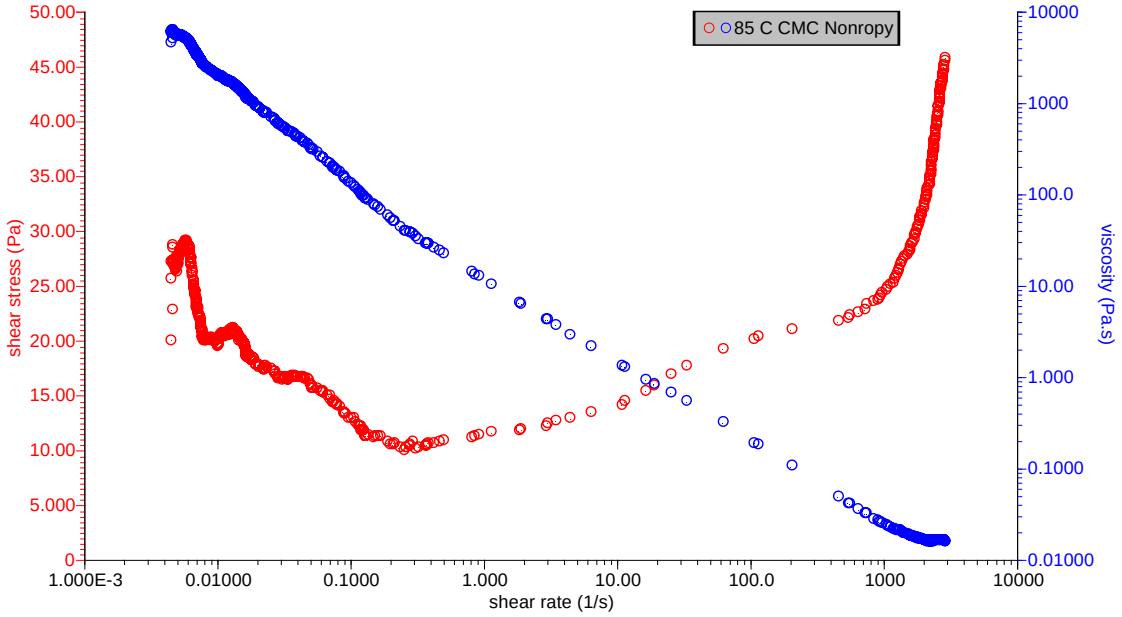


Figure 11 Graphs of Shear Rate (1/s) vs. Shear Stress (Pa) and Shear Rate (1/s) vs. Viscosity (Pa.s) for CMC Nonropy Samples

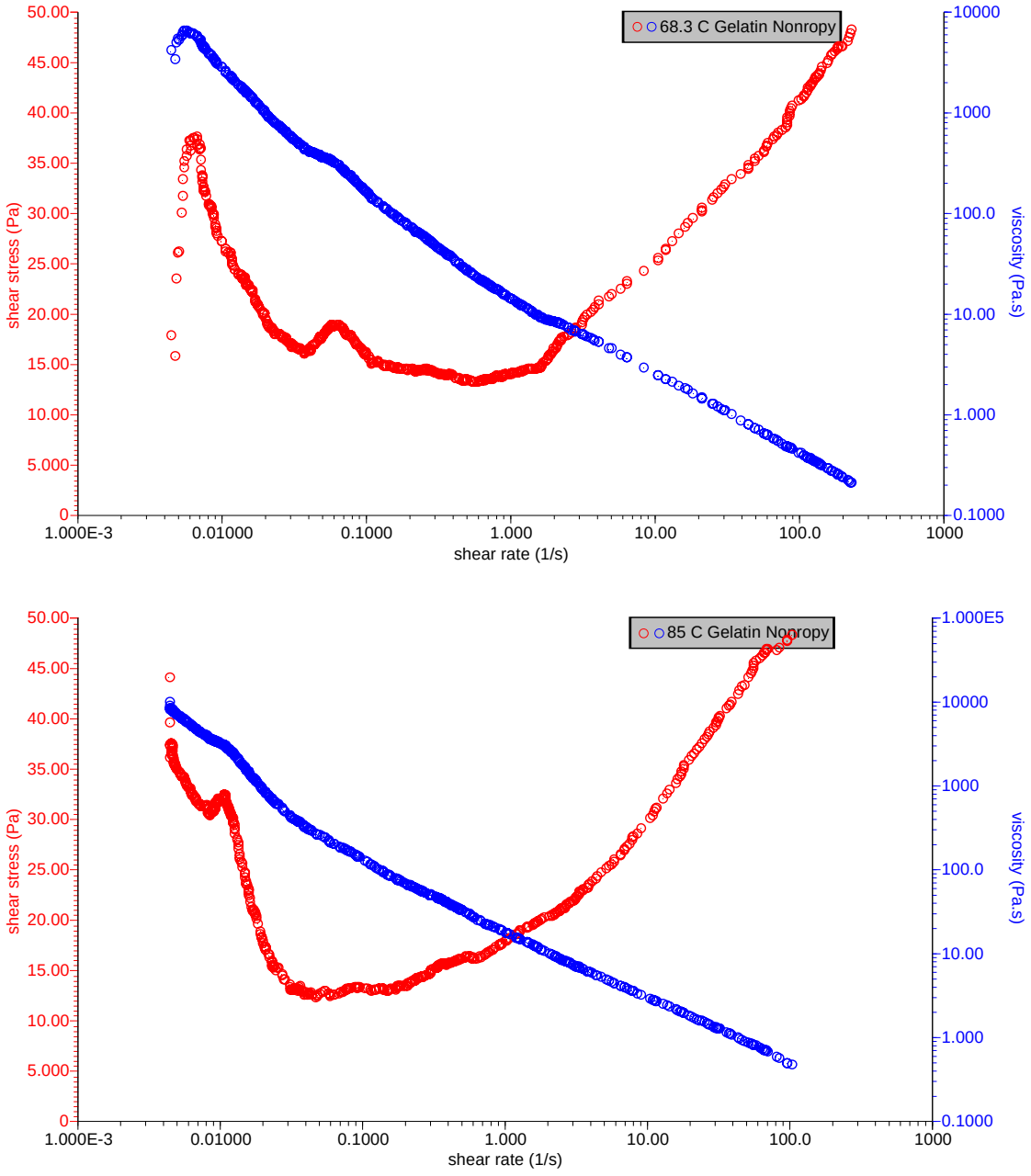


Figure 12 Graphs of Shear Rate (1/s) vs. Shear Stress (Pa) and Shear Rate (1/s) vs. Viscosity (Pa.s) for Gelatin Nonropy Samples

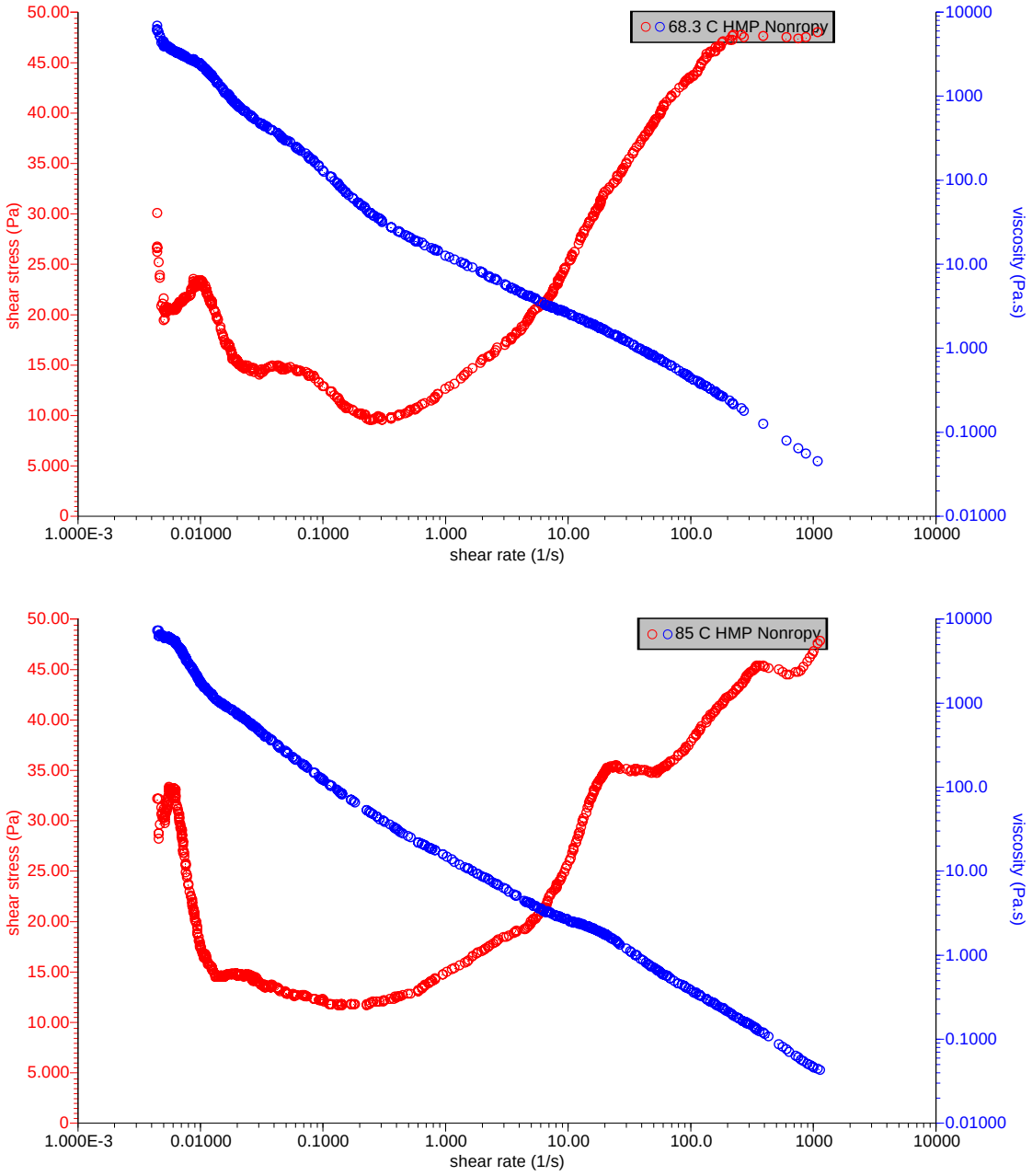
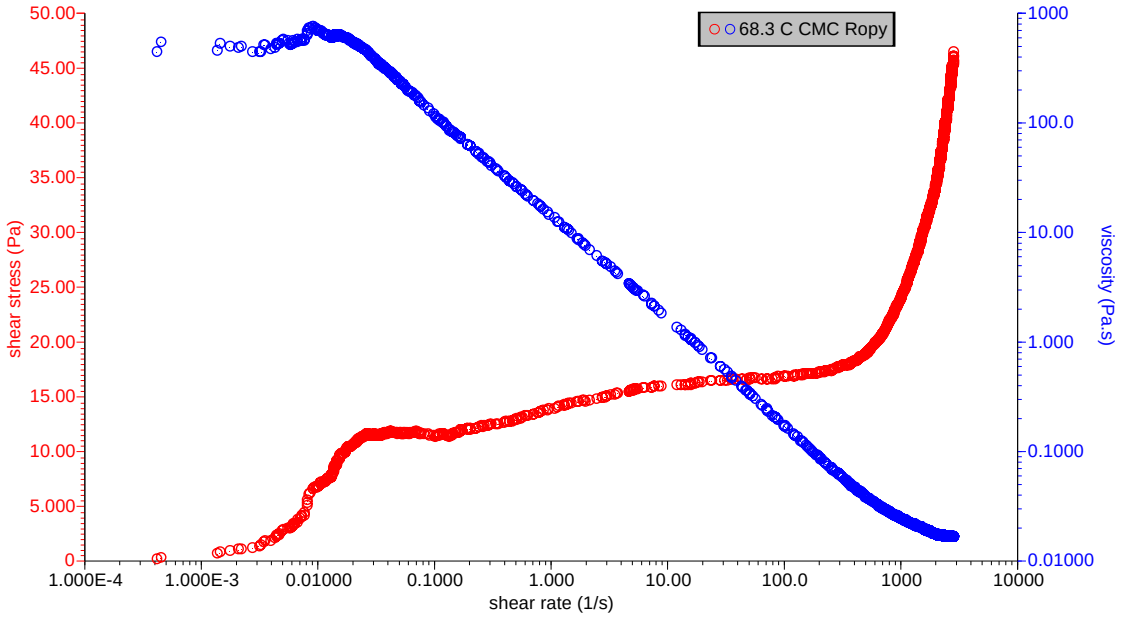


Figure 13 Graphs of Shear Rate (1/s) vs. Shear Stress (Pa) and Shear Rate (1/s) vs. Viscosity (Pa.s) for HMP Nonropy Samples



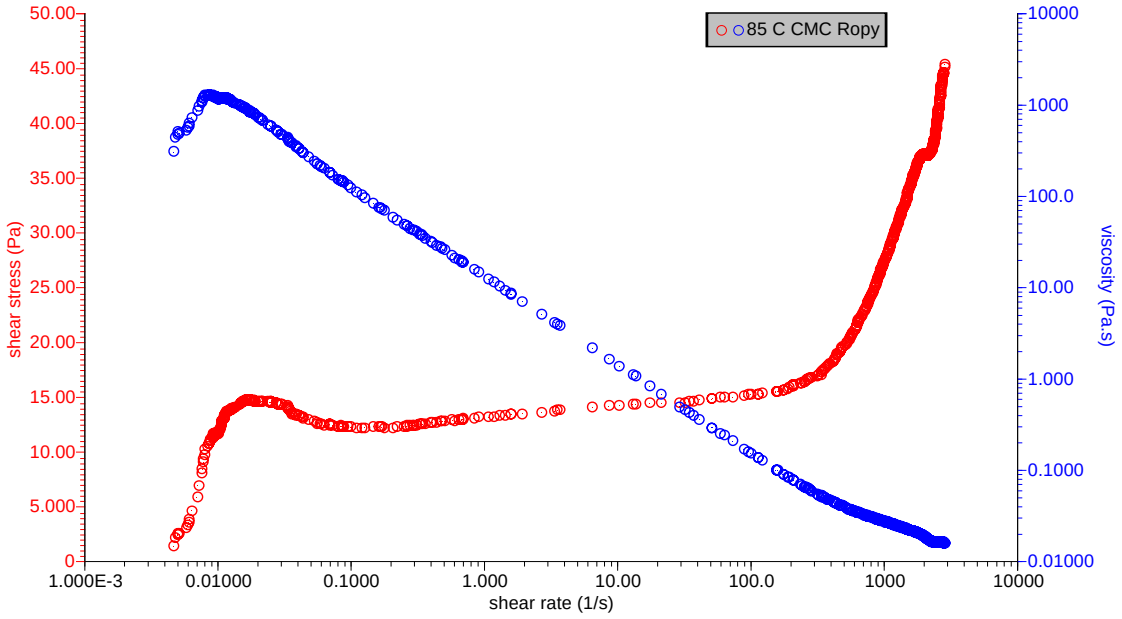
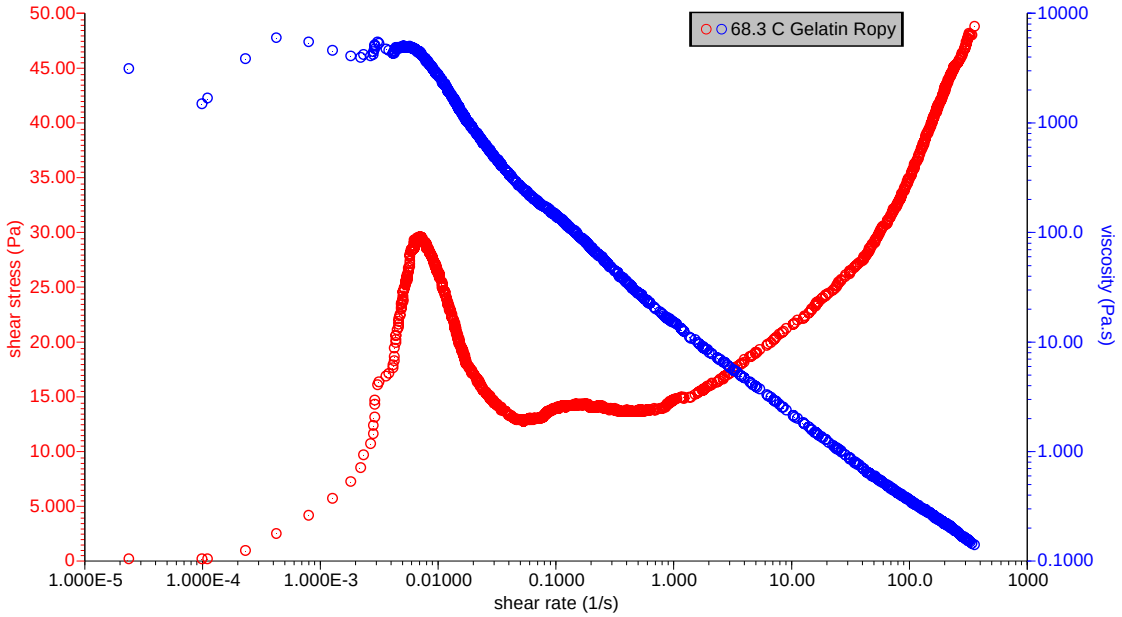


Figure 14 Graphs of Shear Rate (1/s) vs. Shear Stress (Pa) and Shear Rate (1/s) vs. Viscosity (Pa.s) for CMC Ropy Samples



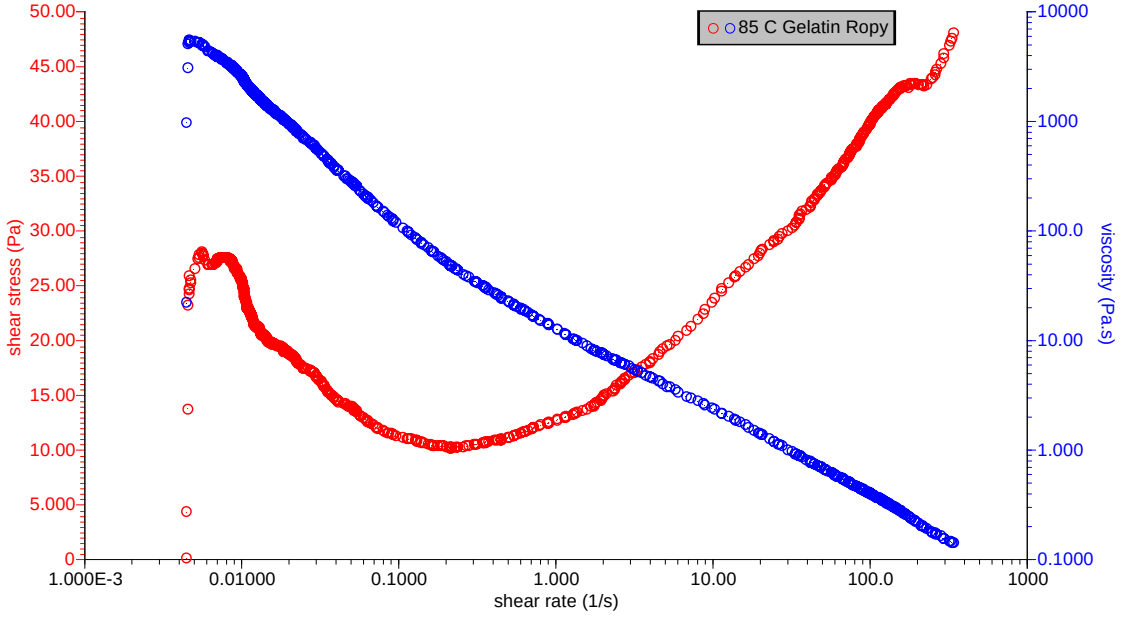


Figure 15 Graphs of Shear Rate (1/s) vs. Shear Stress (Pa) and Shear Rate (1/s) vs. Viscosity (Pa.s) for Gelatin Ropy Samples

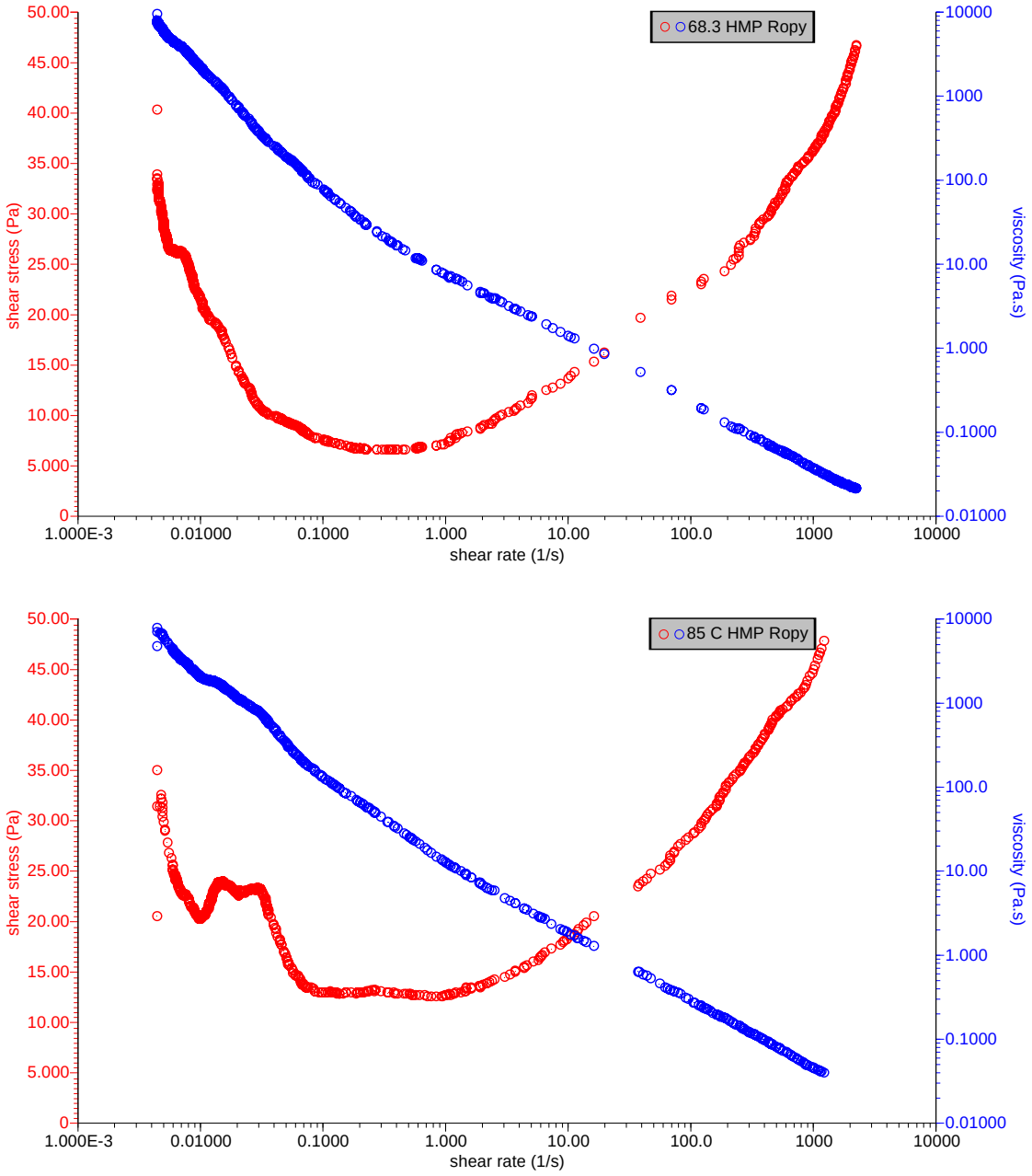


Figure 16 Graphs of Shear Rate (1/s) vs. Shear Stress (Pa) and Shear Rate (1/s) vs. Viscosity (Pa.s) for HMP Ropy Samples

Table 7 Mean Scores for the Rheology Measurements Collected on Drinkable Yogurt Samples at 5 °C.

Treatment			Consistency coefficient (Pa.s)	Rate Index
Past. Temp. (°C)	Stabilizer	Culture		
68.3	Gelatin	Nonropy	23.01	0.077
85.0	Gelatin	Nonropy	24.98	0.013
68.3	CMC	Nonropy	25.01	0.029
85.0	CMC	Nonropy	24.23	0.041
68.3	HMP	Nonropy	23.09	0.104
85.0	HMP	Nonropy	25.01	0.077
68.3	Gelatin	Ropy	23.02	0.086
85.0	Gelatin	Ropy	24.47	0.082
68.3	CMC	Ropy	11.04	0.147
85.0	CMC	Ropy	14.01	0.116
68.3	HMP	Ropy	22.76	0.051
85.0	HMP	Ropy	24.62	0.050

CHAPTER 5
CONCLUSIONS

Both gelatin-stabilized and HMP-stabilized yogurts were more viscous and chalky than CMC-stabilized samples. These yogurt drinks also were the most sour, which agreed with the pH and titratable acidity data. As evidenced in the separation experiment, the stability of the yogurt gel was also maintained fairly well with both of these hydrocolloids. Gelatin and HMP products were higher in protein content than CMC, as well. However, since the CMC-stabilized samples were so different from the other drinkable yogurts produced, comparison between these products is limited. Thus, better parallels can be drawn between the yogurts made with gelatin and HMP. Significant differences were determined in viscosity, with the gelatin samples being thicker, regardless of the type of culture used for fermentation. These differences are also seen in the rheology data. Samples made with HMP were found to be chalkier than those made with gelatin, though the chalkiness was not regarded as objectionable. The sourness of the samples depended on the pasteurization temperature. Despite the sourness and chalkiness, drinkable yogurts produced with either stabilizer were found to be highly acceptable. Overall, samples heated to 68.3 °C and containing gelatin were determined to be the most acceptable, followed closely by the gelatin-stabilized yogurt that had been heated to 85 °C and the HMP samples heated at either temperature. Heat treatment had some effects on the attributes, such as for mouthcoating, though these results often depended on the stabilizers and cultures used. The presence or absence of the exopolysaccharide from the starter cultures also seemed to have influenced many of

the drinkable yogurt properties. These cultures affected viscosity, mouthcoating, and sourness. In addition, pH and titratable acidity were altered. However, depending on the heating temperature, the effects of the starter cultures on the chalkiness were less obvious. Ropy and nonropy cultures had minimal, if any, impact on overall acceptability. Titratable acidity was found to be higher for the products made with the nonropy cultures.

CMC proved to be completely unacceptable as a stabilizer for the yogurt drink (if added prior to fermentation). The resulting product was thin, lacked flavor, and did not maintain the stability of the product. Large aggregates of protein in the samples made the CMC-stabilized yogurt drinks unpalatable. The tendency for whey separation was great for all of the CMC samples, regardless of the starter culture or heating temperature.

The results demonstrate that there are significant effects on mouthfeel and other attributes of a drinkable yogurt system depending upon the stabilizers used, as well as the processing conditions and starter culture selection. Both gelatin and HMP could maintain the stability of the yogurt, as well as produce a palatable and acceptable product for consumption, despite scoring higher values for chalkiness.

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